WESTERN ASPEN SEEDLING PRODUCTION AND ESTABLISHMENT TECHNIQUES FOR FUEL BREAKS AROUND HIGH USE RECREATION AREAS

James T. Fisher

EISENHOWER CONSORTIUM RESEARCH GRANT NO. RM-81-160-CR

(N.M.S.U. Acct. No. 1-528388)

WESTERN ASPEN SEEDLING PRODUCTION AND ESTABLISHMENT TECHNIQUES FOR FUEL BREAKS AROUND HIGH USE RECREATION AREAS

Final Technical Report

SUBMITTED BY

JAMES T. FISHER

New Mexico State University

October 30, 1986

EISENHOWER CONSORTIUM RESEARCH GRANT NO. RM-81-160-CR

(N.M.S.U. Acct. No. 1-528388)

WESTERN ASPEN SEEDLING PRODUCTION AND ESTABLISHMENT TECHNIQUES FOR FUEL BREAKS AROUND HIGH USE RECREATION AREAS

Final Technical Report

SUBMITTED BY

JAMES T. FISHER

New Mexico State University

October 30, 1986

TABLE OF CONTENTS TAB

IV. UNPUBLISHED TABLES TAB, PAGE

PROJECT TITLE:

WESTERN ASPEN SEEDLING PRODUCTION AND ESTABLISHMENT TECHNIQUES FOR FUEL BREAKS AROUND HIGH USE RECREATION AREAS.

I. PROJECT OBJECTIVES:

A) Develop greenhouse and nursery production techniques to optimize seedling dimensions, vigor, and outplanting survival.

B) Mechanical and chemical site preparation techniques for establishing aspen fuel breaks in recreational areas.

C) Determine outplanting success and cost of container-grown seedlings versus bare-root seedlings for establishing aspen fuel breaks.

A. SEEDLING PRODUCTION

(1) Expt. 1: Aspen Seedling Growth in Hardwood Containers.

Objective: Determine the optimum schedule for production of aspen seedlings having similar proportions but different volumes.

Results of this experiment were reported at the national meeting of the American Society for Horticultural Science as follows:

Erhard, L.A. and J.T. Fisher. 1982. Effect of container size and harvest date on the growth of quaking aspen (<u>Populus tremuloides</u> Michaux.) seedlings. HortScience 17:486 (Abst. 101).

Reference the June 1982 progress report for a description of the treatments and design.

Analyses of variance and mean separation techniques were used to detect treatment differences. Tables 1 through 5 summarize these analyses.

Significant differences were detected among the main effects (week of harvest) for shoot dry weight, root dry weight, root collar caliper, and height growth. Container size also significantly affected these four characters, and container X week of harvest interactions were detected for root dry weight, root collar caliper, and height. See Table 1. Results showed that the 68- and 90-in.^{3} containers produced seedlings with greater shoot and root dry weights, root collar caliper, and height than did 11- or 30-in.^{3} containers. See Tables 2 through 5. Seedlings grown in the 11-in.^{3} container were comparable to those in the 30, and those in the 68-in.^{3} were comparable to those in in 90. All seedlings were plantable at 15 weeks.

Table 1. Split-Plot Analysis of Shoot Dry Weight, Root Dry Weight, Root Collar Caliper, and Height Growth. (*=Significant at the .05 level.)

SOURCE OF VARIATION	DF	SHOOT DRY WEIGHT	ROOT DRY WEIGHT	ROOT COLLAR CALIPER	HEIGHT GROWTH
REPLICATION	3	0.665*	0.027	0.639*	136.6*
WEEKS	9	10.062*	0.592*	19.536*	2741.3*
$ERROR_A$	27	0.177	0.013	0.132	21.6
CONTAINER SIZE	3	6.339*	0.251*	7.555*	1687.3*
CONTAINER • WEEKS	27	0.982	0.040*	0.400*	146.8*
$ERROR_B$	90	0.100	0.008	0.118	23.6
	159				

Table 2. Mean Shoot Dry Weight (g).[1]

CONTAINER SIZE (CUBIC INCHES)	6	7	8	9	10	11	12	13	14	15
11	0.019 A	0.052 A	0.102 A	0.103 B	0.241 B	0.262 C	0.391 B	0.551 B	0.894 B	1.030 B
30	0.016 A	0.036 A	0.056 A	0.140 B	0.208 B	0.356 C	0.430 B	0.611 B	0.821 B	1.203 B
68	0.030 A	0.062 A	0.168 A	0.273 A	0.535 A	0.706 B	0.925 A	1.596 A	2.670 A	3.789 A
90	0.031 A	0.074 A	0.169 A	0.265 A	0.518 A	1.010 A	0.819 A	1.461 A	2.758 A	3.446 A

WEEKS FROM SEED

[1] Values with the same letter are not significantly different ($p \leq .05$).

Table 3. Mean Root Dry Weight (g).[1]

CONTAINER SIZE (CUBIC INCHES)	6	7	8	9	10	11	12	13	14	15
11	0.004 A	0.010 A	0.020 A	0.020 C	0.051 B	0.059 C	0.096 B	0.130 B	0.248 B	0.371 B
30	0.004 A	0.008 A	0.013 A	0.024 BC	0.045 B	0.039 BC	0.102 B	0.135 B	0.201 B	0.343 B
68	0.007 A	0.013 A	0.034 A	0.048 AB	0.119 A	0.165 AB	0.173 A	0.340 A	0.532 A	0.959 A
90	0.007 A	0.015 A	0.037 A	0.053 A	0.132 A	0.242 A	0.196 A	0.344 A	0.600 A	0.695 AB

WEEKS FROM SEED

Table 4. Mean Root Collar Caliper (mm).[1]

CUBIC INCH CONTAINERS	6	7	8	9	10	11	12	13	14	15
11	0.9 A	1.1 A	1.4 A	1.4 B	1.9 B	2.0 B	2.2 C	2.6 B	3.2 B	3.4 B
30	0.8 A	1.0 A	1.1 A	1.5 B	1.8 B	2.3 B	2.3 BC	2.6 B	2.9 B	3.4 B
68	0.9 A	1.2 A	1.7 A	2.0 A	2.6 A	2.8 A	3.0 A	3.7 A	4.5 A	5.4 A
90	0.9 A	1.2 A	1.7 A	2.0 A	2.6 A	3.2 A	2.8 AB	3.4 A	4.4 A	4.8 A

WEEKS FROM SEED

Table 5. Mean Height Growth (cm).[1]

CONTAINER SIZE (CUBIC INCHES)	6	7	8	9	10	11	12	13	14	15
11	3.7 A	5.4 A	8.2 A	8.0 B	13.2 B	15.6 B	17.2 B	21.2 B	22.8 B	23.0 B
30	3.0 A	3.9 A	5.2 A	8.8 AB	10.6 B	15.6 B	17.8 B	21.6 B	25.1 B	28.8 B
68	4.0 A	5.5 A	9.9 A	13.3 A	20.9 A	24.2 A	31.1 A	40.0 A	47.5 A	56.5 A
90	3.9 A	5.4 A	9.6 A	13.4 A	21.2 A	28.5 A	28.5 A	33.4 A	48.5 A	58.4 A

WEEKS FROM SEED

MYCORRHIZATION OF *POPULUS TREMULOIDES* UNDER GREENHOUSE CONDITIONS

By

RAJAA KHAZAL ALI, B. S

A Thesis submitted to the Graduate School
in partial fulfillment of the requirements
for the Degree
Master of Science

Major Subject: Horticulture

New Mexico State University
Las Cruces, New Mexico
December 1985

"Mycorrhization of Populus tremuloides Under Greenhouse Conditions," a thesis prepared by Rajaa K. Ali in partial fulfillment of the requirments for the degree, Master of Science, has been approved and accepted by the following:

September 30, 1985

William H. Matchett

Dean of the Graduate School

Chairman of the Examining Committee

September 30, 1985

Date

Committee in charge:

Dr. James T. Fisher, Chairman

Dr. Donald J. Cotter

Dr. Glyn O. Throneberry

Dr. Fred B. Widmoyer

(2) Expt. 2: Inoculation of Containerized Aspen with Forest Soil Containing Mycorrhizae-Forming Fungi.

Objective: Determine feasibility of infecting container-grown aspen seedlings with forest soil containing mycorrhizae-forming fungi.

This experiment is reported in the 1985 M.S. Thesis prepared by Rajaa Ali:

Ali, Rajaa. 1985. Mycorrhization of _Populus tremuloides_ under greenhouse conditions. M.S. Thesis, New Mexico State Univ., Las Cruces, NM.

ACKNOWLEDGEMENTS

The author wishes to express her sincere gratitude to Dr. James T. Fisher for his guidance, encouragement, and helpful suggestions as a major advisor throughout this study.

Appreciation is also extended to Dr. J. Mexal, for his technical advice, to Dr. M. Southward for his assistance in statistical analysis and to Dr. Fred B. Widmoyer, Dr Donald J. Cotter and Dr Glyn O. Throneberry for their helpful suggestions as members of the author's guidance committee.

The author gratefully acknowledges Dr. Donald Lindsey in the Plant Pathology Laboratory at the College of Agriculture and Home Economics of New Mexico State University for technical assistance and the use of essential facilities. Also, thanks to Robert W. Neumann for his help during the final stages of completing this work.

Finally, special thanks to my husband Ahmed and lovely daughter Farah, who provided support, patience, encouragment and love during my study.

VITA

June 7, 1953- Born at Hilla. Iraq

1970-1974- B.S., Mosul University, Mosul, Iraq

1975-1981- Assistant Researcher at State Organization for Soil and Land Reclamation-Department of Soil Microbiology.

1983-1985- Graduate Student, New Mexico State University

PROFESSIONAL AND HONARY SOCIETIES

Iraqi Agriculture. Engineering. Society

PUBLICATIONS

Y. A. Hamdi, M. Abbas. and R. K. Ali. 1978. Response of chickpea to inoculation under Iraq condtions. Tech Bul. No. 63. State Organization of Soils and Land Reclamation Baghdad, Iraq.

Y. A. Hamdi, Rajaa. k. Ali. 1978. Growth of different Rhizobia strains in media containing dibs or mannitol with malt or yeast extract. Tech. Bull. No. 64. State Orgainzation for Soils and Land Reclamation. Baghdad, Iraq.

FIELDS OF STUDY

Major field: Horticulture

ABSTRACT

MYCORRHIZATION OF *POPULUS TREMULOIDES*
UNDER GREENHOUSE CONDITIONS

BY

RAJAA KHAZAL ALI, B.S

Master of Science
New Mexico State University
Las Cruces, New Mexico, 1985
Dr. James T. Fisher, Chairman

This study focused on aspen seedling quality as determined by the presence of mycorrhizae derived from forest soil inoculum. Specific objectives were to determine the effects of different levels of forest soil inoculum on aspen seedling growth and development, and intensity of mycorrhizal infection.

Seedlings were grown in a greenhouse for nine months in a 2:1:1 mixture of vermiculite, peatmoss and volcanic rock to which was added forest soil inoculum at various levels to establish four treatments. Compared to the control treatment (no inoculum added), the 8 and 16

percent by volume levels significantly increased seedling growth, as evidenced by positive effects on all growth parameters. The 8 and 16 percent treatments increased seedling biomass almost 200 percent and 140 percent, respectively. Shoot and root dry weights were not significantly different for control seedlings and those grown in a medium with 8 percent inoculum that was fumigated with methyl bromide before sowing.

The ectomycorrhizae formed under the inoculation treatments were morphologically similar to types previously reported for aspen. No endomycorrhizae were detected under the conditions imposed by the aforementioned experiment, or in second experiment providing a proven source of VAM inoculum applied in the absence of fertilizer phosphorus.

The 8 and 16 percent treatments increased the intensity of infection above controls 120 and 170 percent, respectively. The same treatments markedly increased the infection of third and fourth order lateral roots. The 8 percent level combined with methyl bromide fumigation before sowing did not significantly increase infection above controls. The degree of growth stimulation attributed to treatments seemed in direct proportion to the intensity of infection caused by a given treatment.

Results suggest that forest soil provides an effective means for ensuring the production of

ectomycorrhizal seedlings. Because no endo forms were detected, results of this study offer few insights into the potential role of endo symbionts in improving seedling quality.

TABLE OF CONTENTS

LIST OF TABLES

LIST OF APPENDIX TABLES

PAGE

LIST OF FIGURES

INTRODUCTION

Quaking aspen (Populus tremuloides Michaux) is the most widely spread deciduous forest tree in North America. In the mountainous West, aspen provides watershed protection, wildlife habitat and wood products while enhancing recreational uses. It is often the first species to return to a disturbed or heavily logged area and thus protects the site until more tolerant species become established. Because aspen has a lower wildfire potential than southwestern conifer types, it has been identified as a fuelbreak species. Wildfires spreading from high elevation conifer forests have been observed to die out in aspen, and healthy stands are regarded by fire managers as relatively fireproof areas.

Through research sponsored by the Eisenhower Consortium since 1982, NMSU researchers have developed techniques for growing western aspen seedlings and for establishing fuelbreaks on prepared sites. The research reported here examined the potential benefits to be gained by providing a source of native mycorrhizal inoculum to container grown seedlings. The need for research on aspen mycorrhizal relationships is based upon the dependence of tree seedlings on fungal symbionts during transplant establishment, and the weakly documented ability of aspen to derive benefits from both ecto- and endomycorrhizae.

REVIEW OF LITERATURE

<u>The Occurrence and Natural Regeneration of Western Aspen</u>

The geographic range of western aspen extends from the mountains of Mexico to northern Alaska. It grows at elevations ranging from sea level to 3,300 m. Aspen appears to be ideally adapted to the cool, relatively dry summers and winters with abundant snow. In aspen stands, summer temperatures above 32 C (90 F) are rare and winter temperatures below -18 C (0 F) are common. Annual precipitation ranging from 40 cm (16 in.) to over 100 cm (40 in.) is accumulated in deep winter snow packs. Aspen is usually confined to small isolated stands or rather narrow transitional zones between conifer forests and grasslands.

The adaptive flexibility of aspen has been attributed to its ability to reproduce asexually from root suckering (Milton and Grant, 1980). Because aspen has a greater lateral root spread than other species, root suckering permits it to advance into open areas where conditions may be unfavorable for conifer seedling establishment. Lateral roots from one individual initiate many shoots producing a clonal stand composed of aspen ramets (Barnes, 1969). Suckers are observed more frequently than collar stump sprouts, which accounted for only 20 percent of stems regenerated from a slashed

five-year old quaking aspen stand (Maini, 1968). The root system is quite contorted, and develops concentrated masses of fine roots in the upper 60 cm of soil (Berndt and Gibbons, 1958). Roots occasionally extend to a depth greater than 2 m.

Although aspen is found on soils ranging from talus slopes to deep, heavy clays, optimum growth is observed on open, porous soils high in calcium, magnesium and potassium. Stand development is poorest on rocky sites which limit the lateral spread of roots, and consequent stand colonization.

Containerized Seedling Production

Within the reforestation context, containerizaton extends the planting season (Aycock, 1974) and provides an obvious benefit where shallow soils prevent nursery stock from developing long roots. In the West, seedling survival has generally been 20 percent greater for container than for nursery stock (Hite, 1974). In Michigan, survival was 87 percent and 50 percent, repectively, for container and nursery grown aspen seedlings (Okafo and Hanover, 1978). In addition, containerization accelerated initial transplant growth. Although more than 100 million container-grown seedlings are produced annually in North America, less than 1

percent of these are hardwoods. However, hardwood seedling production continues to increase as greenhouse cultural methods are developed for new species, and as the demand increase for hardwood products and uses, such as high value veneers.

Greenhouse production provides the opportunity to accelerate hardwood seedling growth through optimal irrigation, fertilization, espacement and photoperiod. Some production facilities also provide carbon dioxide enrichment. Containerized seedlings are produced in months, whereas nursery seedlings routinely require two or three years. Although containerized seedlings cost more than nursery stock, production risks are greatly reduced due to the absence of destructive pests and climatic extremes.

Although containerized hardwoods can be grown to plantable size more rapidly than containerized conifers, special care is required to produce healthy and uniform hardwood crops. Hardwood irrigation is more difficult because 1) leaves shed water, 2) transpiration is high and the need for water replacement is frequent, and 3) thorough rinsing is needed to remove water-applied fertilizers from broadleaves. Hardwoods generally require a larger container than conifers because they concentrate initial growth in large thick tap roots (Tinus, 1974).

Johnson (1974) found that seedling size is a reflection of diameter and volume of the container.

Numerous potting mixtures will produce plantable stock if growers know how to properly use them. Most seedlings succeed with a 1:1 mixture, by volume, of fine grind Canadian sphagnum peat moss and horticultural grade vermiculite, or a 3:2 mixture of medium grind peat moss and vermiculite. Such combinations are lightweight and have excellent cation exchange and water retention properties.

The Role and Importance of Mycotrophy

Marks and Kozlowski (1973); Hacskaylo (1971) and Trappe (1977) noted that 95 percent of the world's vascular plants belong to families that are mycorrizhal. Under conditions of poor soil fertility, mycorrhizae enable woody plants to absorb sufficient amounts of essential soil nutrients to become established. Mycorrhizae are particularly beneficial in promoting phosphorus (P) uptake from P-deficient soils. Infected plants absorb and accumulate more P and grow dramatically faster (Gray and Gerdemann, 1967; Sanders and Tinker, 1971; Hayman and Mosse, 1972; Mosse, 1972; Mosse *et al*., 1973; Rhodes and Gerdeman, 1975; Daft and Hacskaylo, 1977). The ability of mycorrhizal plants to take up more

P has been attributed to the extension of the zone from which P is absorbed by emanating fungal hyphae (Gerdemann, 1975), greater root absorption efficiency, and possibly solubilization of soil P by the fungus (Smith, 1974).

Because afforestation attempts commonly fail due to absence of natural inoculum (Hatch, 1936; McComb, 1943; Shemankhonova, 1962; Mikola, 1973; Marx et al., 1978), nurserymen continually seek methods to encourage mycorrhization through artificial means. Numerous studies have addressed the benefits to be gained from infecting hardwood seedlings with mycorrhizae-forming fungi (Fassi and Fontana, 1969; Theodorou and Bowen, 1970; Theodorou, 1971; Vozzo and Hacskaylo, 1971; Lamb and Richards, 1974; Marx, 1979). In this vein, inoculations with Pisolitus tinctorius have greatly accelerated the growth of nursery grown pecan (Carya illionoensis) seedlings (Marx, 1979) and greenhouse propagated yellow poplar cuttings (Navratil and Rochon, 1981). Similarly (vesicular arbusicular) mycorrhizae have stimulated the growth of sweetgum (Liquidambar styraciflua L) (Schultz et al., 1979; Kormanik et al., 1981).

Reports on aspen mycorrhizal relationships are limited. Vozzo ad Hacskaylo (1974) successfully developed ectomycorrhizae on P. tremuloides by inoculating the seedling medium with soil from a pine-aspen stand where

aspen was the dominant species. According to Malloch and Malloch (1981), _P. tremuloides_ roots commonly exhibit both ecto- and endomycorrhizas. They suggested that the widespread occurrence of the genus _Populus_ may be attributed to its rare ability to form both ecto- and endomycorrhizal associations.

The Production of Mycorrhizal Tree Seedlings

Container seedlings grown in soilless media receiving frequent and high fertilization rates generally lack mycorrhizal development (Molina, 1980). Recognizing the need for infection, nurserymen are increasingly taking steps to provide inoculum and to minimize the adverse impacts of intensive culture on fungal colonization. The successful production of mycorrhizal seedlings is contingent upon type and age of inoculum used, timing of inoculation, inoculum density, inoculum placement in the growing medium, and a number of host and fungal interactions (Maronek _et al_., 1981). Mycorrhizal inoculation can be performed before or during seed sowing, or after seedling emergence. The most efficient time to inoculate is when seeds are sown. An efficient time to inoculate cuttings is at the time of placing cutting in it propagation bud.

In developing countries, soil or humus collected from established pine plantations is routinely used as a source of mycorrhizal inoculum (Mikola, 1973). Nurserymen having access to more advanced techniques avoid contaminating seedlings with soil-borne pathogens by introducing a specific fungal symbiont to nursery or container stock via basidiospores or pure vegetative mycelia. *Pisolithus tinctorius* spores mixed with moistened vermiculite, kaolin or sand have been successfuly used to infect nursery and container grown southern pines (Marx *et al*., 1976; Marx *et al*., 1978). However, the vegetative inoculum approach receives greater attention and has been repeatedly recommended (Bowen, 1965; Marx, 1980; Mikola, 1973; Shemankhonova, 1962; Trappe, 1977). Researchers continue to develop more efficient techniques to minimize the amount of vegetative inoculum required to obtain the level of root infection necessary to improve transplant performance.

Endomycorrhizal infections have been successfully established following plant inoculation with soil containing spores, infected roots and symbiont hyphae or spores mixed with a moistened carrier such as vermiculite. However, infection with soil inoculum is considered to be more rapid than from spore inoculum (Ferguson, 1981). For example, sudan grass roots were more rapidly infected when soil inoculum was used

compared to spores (Hall, 1976). This was true especially at low spore density. Plant size is often associated with post-infection sporulation because plants with large, extensive root systems allow greater mycorrhizal colonization than plants with smaller root mass (Daft and Nicolson, 1972; Saif and Khan, 1977).

Seedling fertilization remains a key issue in establishing and promoting the growth of the fungal symbiont. Kormanik et al. (1977) and Schultz et al. (1979) showed that mycorrhizal sweetgum seedlings grew equally well when soil-extractable P ranged from 8 to 45 ppm. However, Yawney et al. (1982) concluded that non-inoculated and Gigaspora margarta-infected sweetgum seedlings grew best in a soil having a pH of 4.5 and containing 100 ppm of P, after subjecting seedlings to soil pH values of 4.5 to 7.8 and soil P concentrations from 25 to 100 ppm.

Because mycorrhizal root systems are able to adequately supply P at low soil P concentations, nitrogen (N) availability may be more limiting and this may explain why the optimal P level varies from one study to another. For instance, Brown et al. (1981) showed that soil P levels of 7-11 ppm were high enough to produce large mycorrhizal seedlings, but that satisfactory growth occurred only when adequate nitrogen (N) was available. Similarly, Konckeki and Read (1976) showed that Festuca

ovina L growth was stimulated by increasing soil P levels only when nitrogen N was present in sufficient amounts. High P levels clearly inhibit mycorrhizal infection (Menge *et al.*, 1978; Ratnayake *et al.*, 1978). It follows that granular fertilizers which are incorporated into a planting medium should contain micronutrients but little or no P. A coarse sand fertilized with a modified Hoagland's solution lacking P can provide a planting medium conducive to the development of mycorrhizal symbiosis.

Conclusions Drawn from the Literature Review

Clearly, the root system is the key to aspen silviculture and natural regeneration management in the southern Rockies. Moreover, the ability of aspen to produce vegetative shoots provides an unusual opportunity for gaining nature's assistance in expanding the reaches of a planted stand.

The production of containerized aspen seedlings will require containers suited to hardwoods and appropriate irrigation and pest management strategies. Containerized aspen seedlings may have higher survival and initial growth potential than nursery stock.

Hardwoods as well as conifers receive substantial, if not obligatory, benefits from their mycorrhizal associations. The adaptive flexibility of aspen may

reside in its ability to establish both ecto- and endomycorrhizal symbioses.

Successful aspen mycorrhizal inoculation will require a suitable source of inoculum that must be applied under appropriate host and cultural conditions.

OBJECTIVES

The objectives of this study were to determine the following:

1. The effect of different levels of soil mycorrhizal inoculum on aspen seedling growth;

2. The effect of the mycorrhizae on growth of the root system of aspen seedlings; and

3. The effect of low and high levels of P on the development of VA mycorrhizae.

MATERIALS AND METHODS

Experiment 1: Aspen Seedling Response to Soil Inoculation Treatments.

Experiment 1 was conducted to determine the effect of different levels of inoculum on aspen seedling growth, root development and intensity of mycorrhizal infection. Greenhouse studies examined the effects of inoculation on seedlings grown under standard production greenhouse conditions with the exception of fertilization treatment. Laboratory studies examined the effect of inoculation on root infection and development as determined by accepted procedures (Giovannetti and Mosse, 1980).

A. Greenhouse studies

Aspen plants were grown in pots containing by volume 50 percent peatmoss, 25 percent vermiculite and 25 percent volcanic rock mixed with different levels of inoculum to form 4 experimental treatments. Treatments included 8 percent soil, 16 percent soil, 8 percent soil with methyl bromide application for a minimum of 48 hours before sowing, and the control without inoculum. Sixty "Styroblock" containers, 20 cm in length, 5.1 x 5.1 cm wide and deep, were used. Each container had 30 cavities, which were filled with inoculated media (Figure 1). Each container was sterilized in commercial bleach

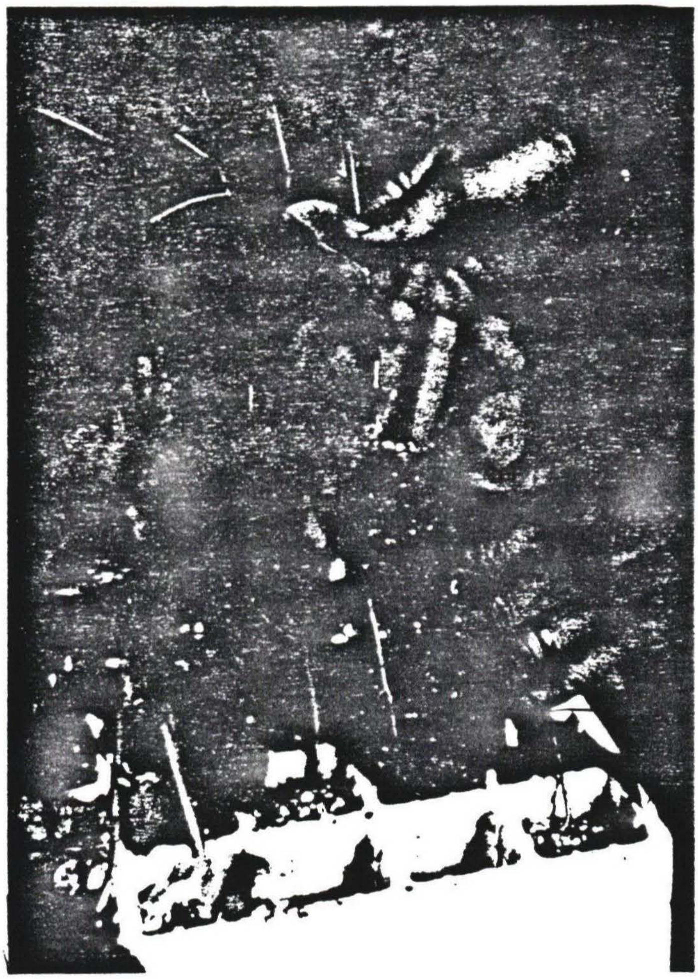


Fig 1. Styroblock container showing the root system of aspen (P. tremuloides) after 9 months under greenhouse conditions

(10 percent solution of sodium hypochlorite) and rinsed with water before being filled with medium.

Soil inoculum was collected from beneath aspen trees at five points within a 200 x 200 m area about 16 miles east of Santa Fe, New Mexico. The elevation of the collection site is 2700 m. The soil and root particles passing through a 0.5 x 0.5 cm mesh screen were mixed thoroughly with the medium on the the same day (Table A1.).

Aspen seed used in this experiment were obtained from Lake City, Colorado, in 1982 and stored in coin envelopes over Drierite at -2 C until needed. Seeds were separated from seed hairs (the "coma" or "cotton") using the vacuum cleaner screening procedure described by Roe and McCain (1962). Seed germination capacity was determined before experimental sowing by placing 20 seeds on moistened filter paper in a Petri dish. Vigorous seeds usually germinate within 48 hours.

Because the aspen seed is very small, the seed were initially sown in a transplant tray filled with a mixture of vermiculite, peatmoss and volcanic rock (2:1:1 v/v). After sowing, the seeds were covered with a thin layer of perlite and irrigated twice daily. After two weeks the seedlings were transplanted into Styroblock cavities filled with the basic mix with the amount of soil inoculum specified by treatment. All seedlings were grown

in the greenhouse from September 17, 1983 through June 1984. Greenhouse photoperiod and temperature were in accordance with environmental conditions recommended for growing containerized western conifers under greenhouse conditions (Tinus and McDonald, 1979). Plants were watered twice daily for two weeks, then daily for the next two weeks. One month after transplanting seedlings received Hoagland's complete nutrient solution applied at one-half strength every 4 days.

The experimental design was a randomized block with 4 treatments, 3 blocks and 4 replications. Each replicate had twelve containers, three per treatment. The experimental unit was 10 seedlings randomly assigned for regular growth measuremants.

Five seedlings were harvested from each unit nine months after transplanting and washed free of the growth medium. Leaf area was determined by a Li-Cor leaf area meter.

Root fresh weights were measured to the nearest .01 g. Roots were washed free of medium and blotted with paper towels. Shoot masses were dried at 70 C beginning on the day following harvest. Roots and shoot lengths were measured to the nearest 0.1 cm. Root collars, calipers were measured to the nearest 0.1 mm.

The F test was used to determine significant effects among treatments on seedling growth and infection

intensity. Data analyses were accomplished through the use of the Statistical Analysis System (SAS). The overall value for the various factors was significant at ($p<0.05$).

B. Laboratory work

All root samples were preserved after harvest in FAA (a mixture of 13 ml formalin, 5 ml glacial acetic acid and 200 ml of 50 percent ethanol) and stored at room temperature until evaluated. The presence and influence of mycorrhizae on root development were determined through laboratory examinations.

A lateral root harvested at a random location along the primary root was placed (without staining) under an Awild M5 microscope and the intensity of infection estimated by using the gridline intersect method described by Giovannetti and Mosse, 1980. Essentially, infection intensity is determined by the number of points at which mycorrhizal structures intersect with the gridlines. The "root slide method" was not used to estimate degree of infection because the morphology of an aspen seedling root system disallowed the estimation of infection intensity on a percentage basis.

The lengths of secondary, tertiary and fourth order roots were measured by counting the intersect of any

order of the root with the gridlines. Each grid square represents one centimeter square. The degree of infection has been quantified by dividing the root length by the number of mycorrhizae in each root order.

Measurements of the secondary, tertiary and fourth order root and degree of infection were made under 12x and 50x magnification using a calibrated eye piece. Glycerol was added to keep the roots moist and partially stationary.

Photographs were taken under 200 x magnification. A freezing microtome was used to prepare root cross sections.

To examine the presence or absense of VA mycorrhizae several staining techniques were used (Phillips and Hayman, 1970; Kormanik *et al*., 1980; Karow, 1984). A modified version of the Kormanik *et al*. (1980) method provided the highest resolution of fungul structures and host tissues. The critical alteration was to avoid the use of the autoclave by heating roots at 94 C for 20 minutes in 10% KOH and phenol. Slides of root segments were examined with a compound light microscope.

Data were analyzed using the same procedures described in greenhouse studies.

Experiment 2: The Effect of Growing Medium P Level on VAM Infection

Expt. 2 was conducted to determine why VA mycorrhizae were not detected in Expt.1 (see results). Specifically, the procedure was to examine the role of P availability on VAM infection in the presence of a proven source of VAM inoculum.

Twenty non-inoculated aspen seedlings grown in Styroblocks according to procedures described under Expt.1 were transplanted into 15-cm diameter pots. *Glomus mossea* inoculum obtained from potted sorghum infected isolate in accordance with procedures described by Ferguson (1981) was added to the aspen seedling medium. Inoculum was pipetted into the medium at eight locations equidistant from the seedling root collar (Table A2). Seedlings were watered twice weekly with one-half strength Hoagland's complete nutrient solution lacking P.

After four months, seedlings were harvested and roots were gently washed to remove the growing media. Infection levels were quantified in accordance with procedures described by Kormanik and McCraw, (1982) and the gridline intersect method (Giovannetti and Mosse, 1980).

RESULTS

Expt. 1. Aspen Seedling Response to Soil Inoculation Treatments

A. Treatment effects on seedling shoot growth

The 8 and 16 percent inoculation treatments without methyl bromide significantly increased seedling growth (Fig 2). The 16 percent treatment produced significantly more growth than the 8 percent treatment. Most notable was the impact of 8 and 16 percent treatments on shoot biomass, which was almost two times greater than control seedlings under the 16 percent treatment. The 8 percent level without methyl bromide increased shoot fresh weight 140 percent. Although the 8 percent (without methyl bromide) and 16 percent inoculation levels creased more favorable plant response than the control and methyl bromide treated seedlings, leaf area and leaf number per seedling were not significantly different between the inoculated and noninoculated seedlings (Table 1).

B. Treatment effects on root growth

The 16 percent treatment increased root dry weight almost 140% (Fig. 3). The 8 percent treatment produced a slight increase (106%) but when soil was treated with methyl bromide actually growth reduced by 12 percent as

Table 1. Aspen seedling growth responses to inoculation and control treatments after 9 months under greenhouse condition (Expt. 1)

Treatment	Shoot Height (cm)	Root length (cm)	Fresh wt. (g/plant) Shoot	Fresh wt. (g/plant) Root	Dry wt. (g/plant) Shoot	Dry wt. (g/plant) Root	Leaf No.	Leaf area	Caliper
16% soil	34.58a	22.87a	5.06a	11.95a	1.85a	2.24a	21.42a	93.72a	43.83a
8% soil	27.27a	21.48b	3.64b	9.58b	1.27b	1.64b	22.13a	77.2ab	39.63b
8%s + M.B.	25.28b	20.14c	3.15b	8.94b	1.07c	1.40b	17.10a	64.01b	38.92b
control	26.83b	19.86c	2.60c	8.00c	0.99c	1.53b	20.97a	66.77b	38.18b

Means with the same latter are not significantly different at 5% level of probability as determined by LSD.

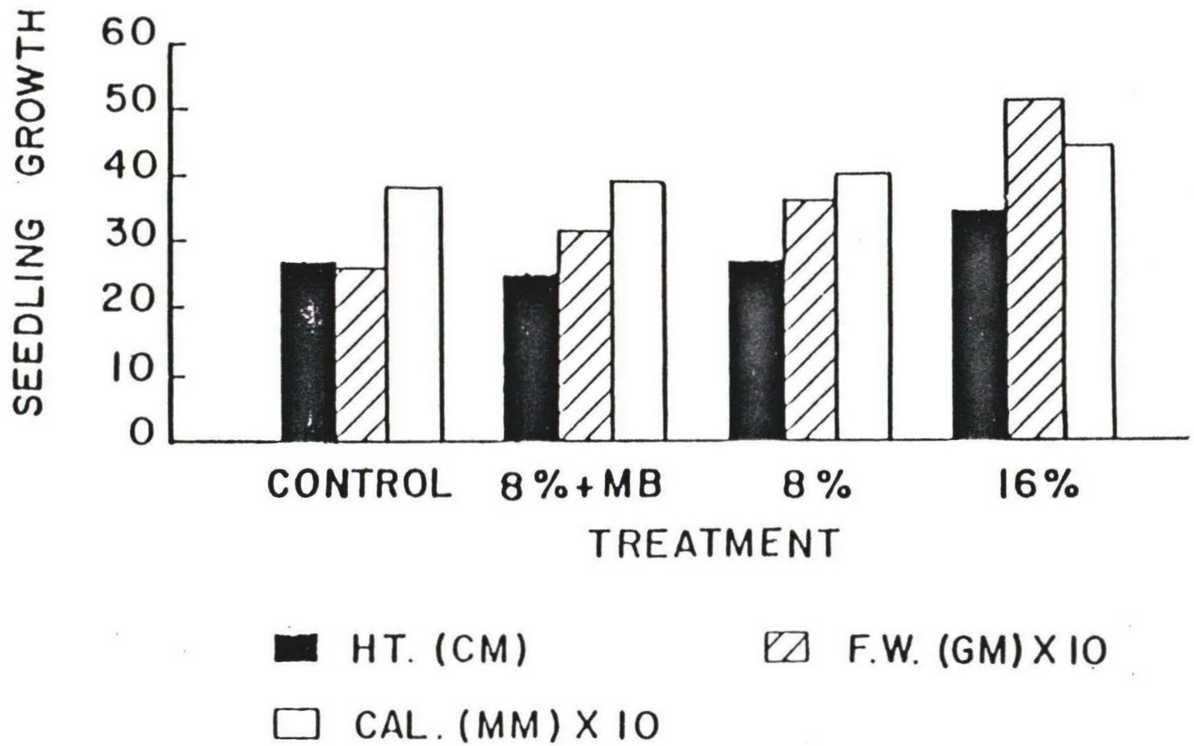


Fig 2. The effect of inoculum treatments and control on seedling growth of aspen

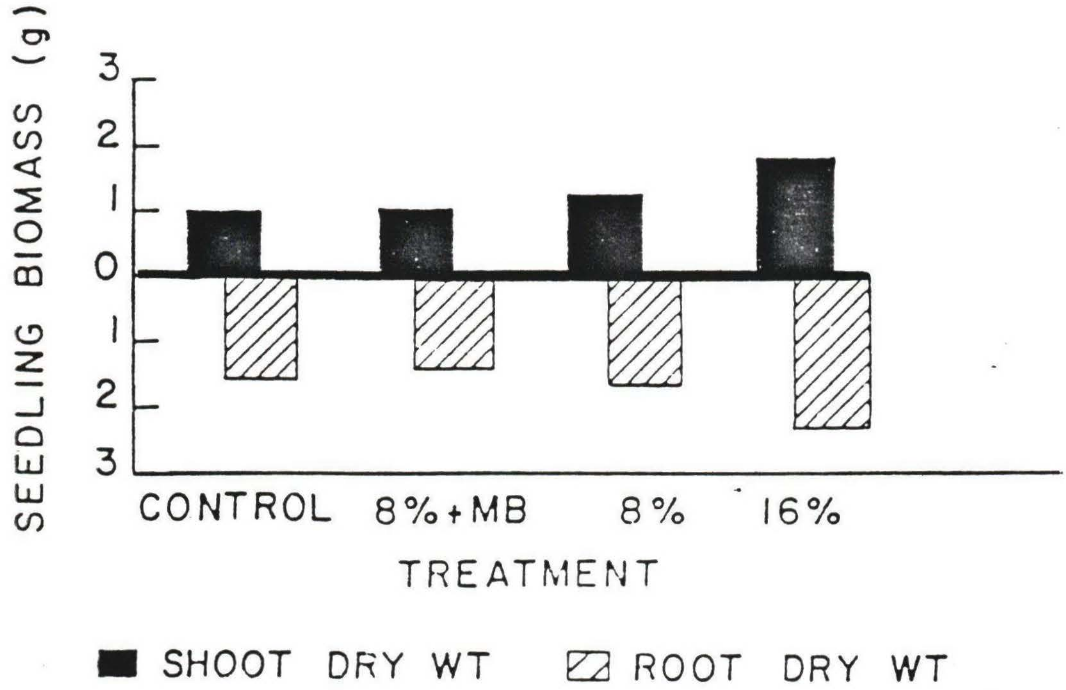


Fig 3. The effect of inoculum treatments on roots and shoots biomass of aspen after 9 months under greenhouse condition (Expt.1)

compared to controls. The mean rate of root elongation was significantly greater for inoculated seedlings than for noninoculated seedlings (Table 1). Roots of noninoculated seedlings were significantly shorter and weighed less than inoculated seedlings (16 percent treatment). However, the mean length of secondary roots was not significantly different among the treatments. The morphology of root system showed no differences between the treatments.

C. Treatment effects on the development of ectomycorrhizae

The ectomycorrhizae formed in the inoculated treatments were morphologically similar to those described by Vozzo and Hacskaylo (1974) on *Populus tremuloides*. Ectomycorrhizae were thick, straight and unbranched. Black hyphae radiated from the mycorrhizal surface with glabrous mycorrhizae occurring primarily along the length of the roots. Ectomycorrhizal roots formed swollen apicies, and mycelial hyphae observed on the apical mantle varied from 300 μm to less than 20 μm in diameter. In addition the mycelial mantle surrounding the epidermal layer emanated from an intercellular network of hyphae among cortical cells forming a Hartig net. No sporophores were observed in inoculated or control cavities. The hyphae extended externally in all

directions into the roots of the growing medium. Inoculated roots showed well-developed ectomycorrhizae, evidenced by mantles which were easily detected microscopically (Figures 5, 6). The non-mycorrhizal roots appeared long, straight and uniform in diameter (Fig 7). Ectomycorrhizae were observed on roots of all treatments. Noninoculated seedlings were apparently infected with inoculum indigenous to the greenhouse. However, the incidence of infection was much greater for 8 percent (without methyl bromide) and 16 percent treatments, and seedling growth appeared closely related to level of infection. Seedlings with ectomycorrhizae had significantly greater heights and root and shoot biomass than controls and methyl bromide treated seedlings, which showed much less infection.

The 8 percent (without methyl bromide) and 16 percent treatments essentially increased the intensity of infection above controls 120 and 170 percent, respectively (Fig. 4 and Table 2). Seedlings subjected to the 8 percent (without methyl bromide) and 16 percent treatments had six times more infected third order laterals and almost nine times more infected fourth order laterals. The 8 percent plus methyl bromide treatment did not significantly increase infection above controls.

Table 2. Treatment effects on primary root growth and ectomycorrhizal infection intensity (Expt. 1).

Treatment	Lateral Root(cm)	Length(cm) of root per plant*			Number of infections*		
		$2^{o}L$	$3^{o}L$	$4^{o}L$	$2^{o}L$	$3^{o}L$	$4^{o}L$
16% soil	15.48a	37.43a	44.52a	20.82a	12.10a	34.57a	24.50a
8% soil	13.43a	34.60a	45.12a	17.23ab	8.75ab	37.18a	22.75a
8%s + M.B.	14.57a	33.75a	36.85a	11.75ab	4.87bc	4.87b	3.48b
control	14.83a	14.42a	35.95a	6.68b	3.90c	5.28b	2.62b

* $2^{o}L$= second order roots, $3^{o}L$= third order roots, $4^{o}L$= fourth order roots

Means with the same letter are not significantly different at 5% level of probability as determined by LSD.

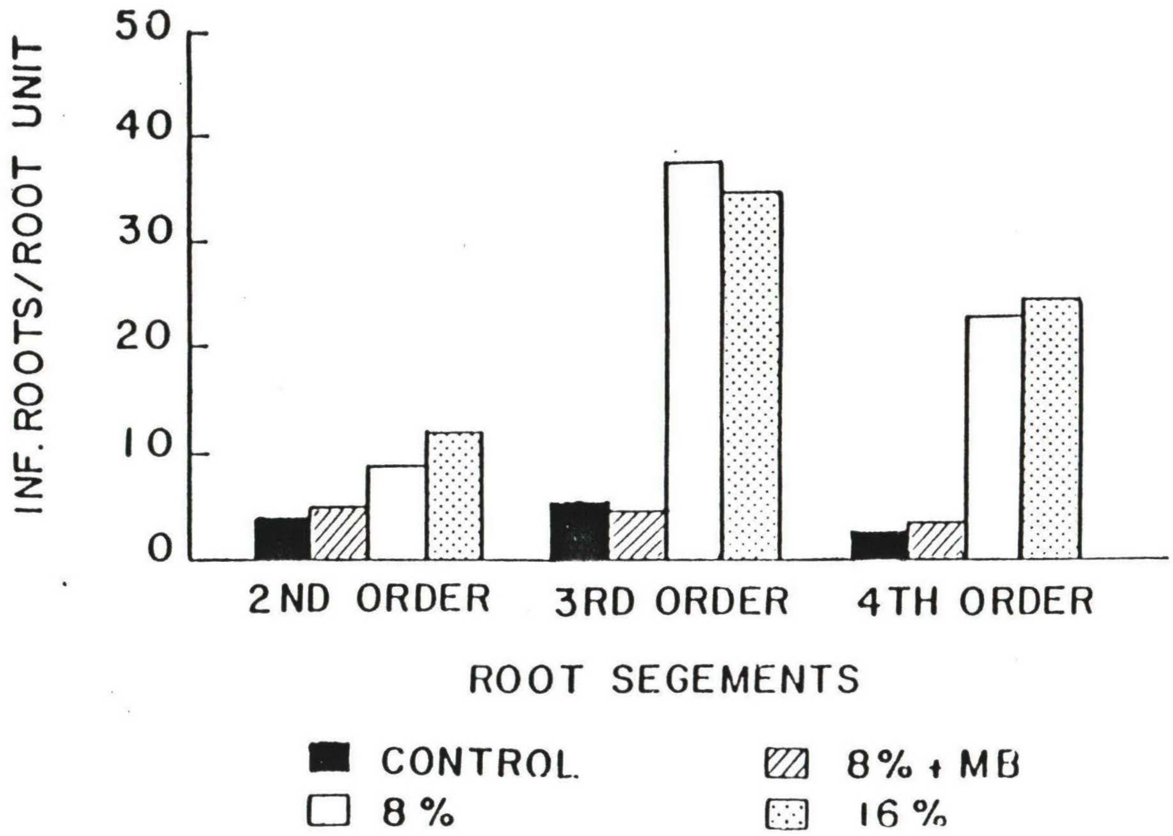


Fig 4. Treatment effects on ectomycorrhizae infection intensity among 2nd, 3rd and 4th-order roots of containerized aspen seedlings (Expt.1)

Fig 5. Different views of ectomycorrhizae apex with fungal mantle, and darker meristematic region (75x magnification) (Expt. 1)

Fig 6. Fungal mantle showing septation near the base of infected aspen root (1000x magnification)

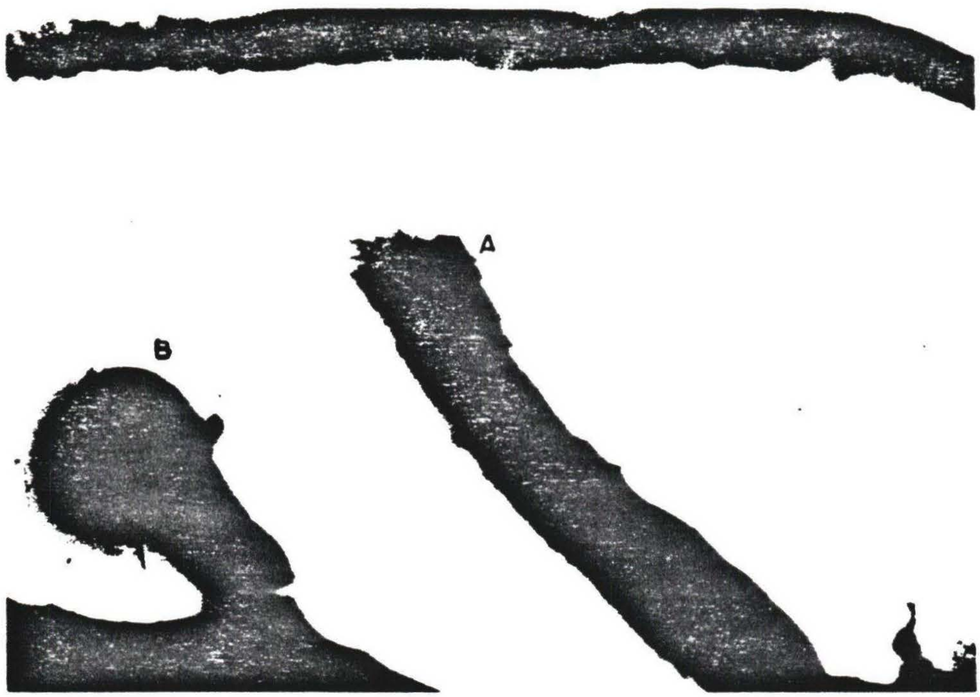


Fig 7. General view of aspen showing the non-infected (A) and infected roots (B)

D.Treatment effects on endomycorrhizal development

Extensive histological examinations did not detect the presence of aspen seedling endomycorrhizae. No vesicles or arbuscles were observed in stained sections obtained from primary, secondary, third and fourth order roots.

Experiment 2. Effect of Growing Medium P Level on VAM Infection

No VAM infections were detected indicating that the presence of a proven source of VAM inoculum applied in the absence of ferilizer P did not result in infection.

DISCUSSION AND CONCLUSIONS

The positive effects associated with soil inoculation treatments applied in this study essentially agree with results reported for other hardwood species (e.g., Clark 1963, 1964). The poor growth obtained without soil inoculum adds credence to the belief that mycorrhizae play a significant role in *Populus* growth and occurrence, as suggested by Malloch and Malloch (1981).

Among the significant relationships determined in this study is that seedling growth is promoted in proportion to the intensity of ectomycorrhizal infection. Also noteworthy are results indicating that increased amounts of inoculum in the growing medium result in faster root colonization, as suggested by Furlan and Fortin (1973).

The positive effects of inoculation and ectomycorrhizal infection observed in this study may be attributed to several factors. One could surmise that infection increased nutrient absorption capacity and provided some protection against pathogenic organisms that routinely challenge greenhouse production efforts. Additionally, infection may alter seedling hormonal balances, which may have greater impacts than nutrient uptake effects (Safir *et al.*, 1971 and 1972).

The methyl bromide treatment was applied primarily to separate mycorrhizal effects from those derived changes in the physical and chemical properies of the growing medium caused by the addition of forest soil. Because methyl bromide produced more negative than positive effects, the use of the fumigant as applied in this study is questionable at best.

Because extensive histological examinations failed to detect endomycorrhizas, I am forced to consider that at least one of the following statments is true:

1) endomycorrhizal inocula were absent in the native soils sampled; 2) greenhouse or seedling conditions prevailed against endomycorrhizal infection; 3) aspen is primarly ectomycorrhizal in habit.

Because Expt. 2 failed to detect VAM mycorrhizae under no fertilization, it seems reasonable that the absence of infection was not directly due to the level of P applied in Expt.1. In addition, Kormanik (1980) grew high quality VAM-infected yellow poplar containerized seedlings supplied with 25-30 ppm P. In my study 15-30 ppm were available, so it seems more reasonable to suspect that the presence of viable ectomycorrhizal inoculation, confirmed by the responses and infections observed, supports the view that forst soil collections were handled appropriately and that VAM inoculum was not destroyed if present. This leaves open the view that aspen may, infact,

be primarialy ectomycorrhizal in habit as suggested by the report submitted by Vozzo and Hacskaylo (1974).

Commerical nurserymen and horticulturists may be able to significantly increase the size and plantability of containerized aspen seedlings by promoting ectomycorrhizal infection and development. Several methods could be used to ensure infection, including the use of vegetative mycelial cultures, or cultures obtained from the production of inoculum. However cultures obtained through the latter process may become contaminated with other microorganisms (yeast, bacteria, and fungi) or insect pests.

In this vein, the use of inoculum obtained from beneath aspen stands may provide unique benefits. Inoculum obtained from forest stands probably contains several species of ectomycorrhizal fungi. This raises the probability that at least one effective symbiont will be present in the soil inoculum. It is also more probable that seedlings planted in the area from which the inoculum was obtained will be infected by a fungal species adapted to the reforestation site. Certainly the soils beneath aspen stands provide a readily accessibble store of vast amounts of inoculum that would require considerable effort to reproduce artifically.

The positive effects of ectomycorrhizal infection observed in this study suggest that mycorrhizae may

greatly benefit members of the genus *Populus*, as suggested by Malloch and Malloch (1981) .

However, results further suggest that these advantages may not offset the hazards associated with potential introduction of pathogenic organisms present in forest soils. Clearly research is needed to determine the comparative benefits associated with inoculum provided by pure cultures.

The following conclusions are drawn from the results obtained in this study :

1) Under the fertilization conditions imposed on seedlings grown in Styroblock cavities, the 8 and 16 percent soil inoculation treatments significantly increased seedling growth and ectomycorrhizal infection. Because these effects have been positively related to seedling quality, it is concluded that the use of soil inoculum should improve transplant success.

2) Specifically, the 16 percent treatment is recommended over the 8 percent treatment because the former results is greater seedling growth.

3) The positive growth effects obtained from inoculation treatments were clearly related to the intensites of infection attributed to them.

4) Because soil and cultured VAM inoculations did not result in detectable endomycorrhizal infections, the

results of this study does not support the view that aspen is closely associated with VAM symbionts.

LITERATURE CITED

Aycock, O. 1974. Field performance of containerized seedlings in the southern region, p. 321-324. _In_ Proc North Amer. Containerized For. Tree Seedling Symp. Great Plains Agri. Coun. Publ. No. 68.

Barnes, B. V. 1969. Natural variation and delineation of _Populus tremuloides_ and _P. grandidentata_ in northern lower Michigan, Silva. Genet. 18:130-142.

Berndt, H. W and R. D. Gibbons. 1958. Root distribution of some native trees and understory plants growing on three sites within ponderosa pine watersheds in Colorado, USDA For. Serv. Res. Pap. RM-37, 14 p.

Bowen, G. D. 1965. Mycorrhizae inoculation in forestry practices, Aust. For. 29:231-237.

Brown, R. W., R. C. Schultz and P. P. Kormanik. 1981. Response of vesicular-arbuscular endomycorrhizal sweetgum seedlings response to three nitrogen fertilizers, For. Sci. 27:413-420.

Clark, F. B. 1963. Endotrophic mycorrhizae influence yellow poplar growth, Sci. 140:1220-1221.

______ 1964. Microorganism and soil structure affect yellow poplar growth, USDA For. Serv. Res. Pap. CS-9, 12 p.

Daft, M. J. and T. H. Nicolson. 1972. Effect of _Endogone_ mycorrhizae on plant growth IV. Quantitative relationships between the growth of the host and development of the host and the development of the endophyte in tomato and maize, New Phytol. 71:287-295.

_______, and E. Hacskaylo. 1977. Growth of endomycorrhizal and nonmycorrhizal red maple seedlings in sand and anthracite soil, For. Sci. 23:207-216.

Fassi, B. and A. Fontana . 1969. Mycorrhizal synthesis between _Pinus strobus_ (eastern white pine) and _Tuber maculatum_. II. Development of ascocarps, Allionia, 15:115-120.

Ferguson, J. J. 1981. Inoculum production and field application of vesicular-arbuscular mycorrhizal fungi, Ph.D. Thesis, Univ. of Calif, Riverside, 117 p.

Furlan, V. and J. A. Fortin. 1973. Formation of endomycorrhizae by _Endogene calospora_ and _Allium cepa_ under three temperature regimes, Naturaliste Canadien, 100:467-477.

Gerdemann, J. W. 1975. The Development and Function of Roots, p. 575-591. In. J. G. Torry and D. T. Clarkson (eds), Academic Press, New York.

Giovannetti, M. and B. Mosse 1980. An evaluation of techniques for measuring vesicular-arbuscular mycorrhizal infection in roots, New Phytol. 84:489-500.

Gray, L. E. and J. W. Gerdemann. 1967. Influence of vesicular-arbuscular mycorrhizae on the uptake of phosphorus 32 by Lirodendron tulipfera and Liquidambar straciflua, Nature (London). 213:106-107.

Hacskaylo, E. 1971. Mycorrhizae, USDA For. Serv. Misc. Publ. 1189. 255 p.

Hall, I. R. 1976. Response of Coprosma robosta to different forms of endomycorrhizal inoculum, Trans. Br. Mycol. Soc. 67:409-411.

Hatch, A. B. 1936. The role of mycorrhizae in afforestation, J. For. 34:22-29.

Hayman, D. S and B. Mosse. 1972. The role of vesicular arbuscular mycorrhizae in the removal of phosphorus from soil by plants roots, Rev. Ecol. Biol. Sol. 9:463-470.

Hite, W. A. 1974. Container field performance in the Rockies and Plains, p. 306-309. In Proc. North Amer. Containerized For. Tree Seedling Symp. Great Plains Agr. Coun. Publ. No. 68.

Johnson, H. J. 1974. Canadian Forestry Service containerized planting trial in Alberta, Saskatchewan and Manitoba, p. 298-305. In Proc. North Amer. Containerized For. Tree Seedling Symp. Great Plains Agri. Publ. No. 68.

Karow, J. A. 1984. The effect of N, P and K on mycorrhizal infection and growth response of alfalfa, M.S. Thesis, New Mex. State Univ., Las Cruces. 30 p.

Konckeki, H. K. and D. J. Read. 1976. Vesicular-arbuscular mycorrhizae in natural vegetation systems. II. The relationship between infection and growth in Festuca ovin L, New Phytol. 77:655-666.

Kormanik, P. P. 1980. Effects of nursery practices on vesicular-arbuscular mycorrhizal development and hardwood seedling production, Proceedings Southeastern Area Forest Tree Nursery Conf. Lake Barkley, KY. 5 p.

________, and A. C. McGraw. 1982. Quantification of vesicular-arbuscular mycorrhizae in plant roots, p. 37-45. In N. C. Schrenk (ed.), Methods and Principles of Mycorrhizal Research. Am. Phytopath. Soc.

_______, W.C. Bryan and R. C. Schultz. 1977. The influence of endomycorrhizae on growth of sweetgum seedlings from eight mother trees, For. Sci 23:500-506.

_______, _______, and _______. 1980 Procedures and equipment for staining large numbers of plant roots for endomycorrhizal assay, Can. J. Microbiol. 26:536-538.

Lamb, R.J. and B. N. Richards. 1974. Inoculation of pine with mycorrhizal fungi in natural soils. I. Effect of density and time of application of inoculum and phosphorus amendment on mycorrhizal infection, Soil Biol. Biochem. 6:167-171.

Malloch, D and Malloch, B. 1981. The mycorrhizal ecology of two nothern poplars, Proc. Fifth North Amercan Conference on Mycorrhizae (abstract 36 p).

Marks, C. G. and T. T. Kozlowski. 1973. Ectomycorrhizae: Their Ecology and Physiology, Academic Press. New York, 444 p.

Maini, J. S. 1968. Silvics and ecology of <u>Populus</u> in Canad, p. 20-64. <u>In</u>: Growth and Utilization of Poplars in Canada. Can. Dept. For and Rural Develop. Publ. No. 1205.

Maronek, D. M., J. W. Handrix, and J. Kierman. 1981. Adjusting nursery practices for production of

mycorrhizal seedlings during propagation, Proc. Int. Plant Prop. Soc. 31:21-31.

Marx, D. H. 1979. Synthesis of Pisolithus ectomycorrhizae on pecan seedling in fumigated soil, USDA. For. Serv. Res. Note SE-280. 8 p.

_______ 1980. Ectomycorrhizal fungus inoculations: A tool for improving forestation practices, p. 13-71. In P. Mikola (ed.) Tropical Mycorrhizae Research. Oxford Univ. Press, London.

_______, W. C. Bryan and G. E. Codell. 1976. Growth and ectmycorrhizal development of pine seedlings in nursery soil infested with the fungal symbiont, Pisolithus tinctorius, For. Sci. 22:91-100.

_______, W. G. Morris and J. C. Mexal, 1978. Growth and ectomycorrhizal development of loblolly pine seedlings in fumigated and nonfumigated soil infested with different fungal symbionts, For. Sci. 24:193-203.

McComb, A. l. 1943. Mycorrhizae and phosphorus nutrition of pine seedlings in a prairie soil nursery, Iowa Agr. Exp. Sta. Res. Bull. 314:581-612.

Menge, J. A., C. K. Labanouskas. E. L. Johnson, and R. C. Platt. 1978. Partial substitition of mycorrhizal fungi for phosphorus fertilization in the greenhouse culture of citrus, Soil. Sci. Soc. Amer. J. 42:926-930.

Mexal, J. G. and C. P. P. Ried, 1972. The growth of selected mycorrhizal fungi in response to induced water stress, Can. J. Bot. 51:1579-1588.

Mikola, P. 1973. Application of mycorrhizal symbiosis in forestry practice, p. 383-411. In. G. C Marks and T. T Kozolowski (eds), Ectomycorrhizae, Their Ecology and Physiology, Academic Press, New York, 444 p.

Milton, J. B., and C. G. Grant. 1980. Observations on the ecology and evolution of quaking aspen Populus tremuloides in the Colorado Front Range, Am. J. Bot. 67:202-209.

Molina, R. 1980. Ectomycorrhizal inoculation of containerizated western conifer seedlings, USDA For. Serv. Res. Note. PNW-357.

Mosse, B. 1972. The influence of soil type and Endogene strain on the growth of mycorrhizal plants in phosphate deficient soil, Rev. Ecol. Biol. Sol. 9:529-537.

_______, D. S. Hayman and D. J. Arnold. 1973. Plant growth responses to vesicular arbuscular mycorrhizae V. Phosphate uptake by three plant species from P deficient soils labelled with 32 P, New Phytol. 72:809-815.

Navratil, S. and G. C. Rochon. 1981. Enhanced root and shoot development of poplar cuttings induced by Pisolithus inoculum, Can. J. For. Res. 11:844-848.

Okafo, O. A. and J. W. Hanover. 1978. Two-year performance of barerooted and containerized trembling and bigtooth aspen seedlings, Tree Planters' Notes 29:24-28.

Phillips, J. M and J. M. Hayman. 1970. Improved procedures for clearing roots and staining parasitic and vesicular-arbuscular mycorrhizal fungi for rapid assessment of infection, Trans. Br. Mycol. Soc. 55:158-161.

Ratnayake, M. R., T. Leonard and J. A. Menge. 1978. Root exudation in relation to supply of phosphorus and its possible relevance to mycorrhizal formation, New Phytol. 81:543-552.

Rhodes, L. H. and J. W. Gerdemann. 1975. Phosphate uptake zones of mycorrhizal and nonmycorrhizal onions, New Phytol. 75:555-561.

Roe, E. I. and D. P. McCain. 1962. A quick method of collecting and cleaning aspen seed, Tree Planters' Notes 51:17-18.

Safir, G. R., J. S. Boyer and J. W. Gerdenmann. 1971. Mycorrhizal enhancement of water transport in soybean, Science 172:581-583.

________, ________, and ________. 1972. Nutrient status and mycorrhizal enhancement of water transport in soybean, Plant Physiol. 49:700-703.

Saif, S. R. and A. G. Khan. 1977. The effect of vesicular arbuscular mycorrhizae associations on growth of cereals. Part 3, Effect on barley growth, Plant and Soil. 47:17-26.

Sanders, F. E. and P. B. Tinker. 1971. Mechanism of absorption of phosphate from soil by Endogone mycorrhizas, Nature (London) 233:278-279.

Schultz, R. C., P. P. Kormanik., W. C. Bryan. and G. H. Brister. 1979. Vesicular-arbuscular mycorrhizae influence growth but not mineral concentrations in seedlings of eight sweetgum families, Can. J. For. Res. 9:218-223.

Shemankhonova, N. M. 1962. Mycotrophy of Woody Plants, Washington, D. C., US Dept. Commer. (Transl. TT66-51073 (1967)). 329 p.

Smith, S. E. 1974. Mycorrhizal fungi, CRC. Critical Review in Microbiology. 2:273-313.

Theodorou, C. 1971. Introduction of mycorrhizal fungi into soil by spore inoculation of seed, Aust. For. 35:23-26.

_______, and G. D. Bowen. 1970. Mycorrhizal responses of radiata pine in experiments with different fungi, Aust. For. 34:183-191.

Tinus, R. W. 1974. Large trees for the Rockeis and Plains, p. 112-118. In Proc. North Amer. Containerized For. Tree Seedlind Symp. Great Plains Agric. Coun. Publ. No. 68.

_______, and S. E. McDonald. 1979. How to grow tree seedlings in containers in greenhouses, Gen. Tech. Rept. RM-60. Rocky Mountain Forest and Range Experiment station, USDA For. Serv. 210 p.

Trappe, J. M. 1977. Selection of fungi for ectomycorrhizal inoculation in nurseries, Ann. Rev. Phytopathol. 15:203-222.

Vozzo, J. A. and E. Hacskaylo. 1971. Inoculation of _Pinus caribaea_ with ectomycorrhizal fungi in Puerto Rico, For. Sci. 17:239-245.

________, and _______. 1974. Endo and ectomycorrhizal association in five _Populus_ species, Bull. of Torrey Bot. Club. 101:182-186.

Yawney, W. J., R. C. Schultz and P. P. Kormanik. 1982. Soil phosphorus and pH influence the growth of mycorrhizal sweetgum, Soil. Sci. Soc. Amer. J. 46: 1315-1320.

APPENDIX: ADDITIONAL DATA

APPENDIX Table Al. Physical and chemical properties of control and inoculated media used in (Expt.1) *

Treatment	pH	Salt E.C	% O.M	Nitrate N (ppm)	Phosphorus P (ppm)	Potassium K (ppm)
16% Soil	7.22	1.26	16.65	2.30	25.10	45.70
8% Soil	6.93	1.89	21.71	1.60	24.20	55.70
8%S+M.B.*	7.06	1.42	20.90	1.10	14.50	68.20
Control	7.14	1.20	21.78	2.80	30.30	53.10

* Soil inoculum was colllected from beneath aspen tree

* M.B= methyl bromide

Analyses performed at the Soil and Water Testing Laborotary. New Mexiço State University

APPENDIX Table A2. Physical and chemical properties of control and inoculated media (sorghum soil+Glomus mossea) used in Expt. 2.

Treatment	pH	salt E.C	% O.M	Nitrate N (ppm)	Phosphorus P (ppm)	Potasume K (ppm)
16% soil	7.56	3.12	14.06	16.7	4.36	17
8% soil	7.16	1.93	7.03	34.7	6.59	34
8%S+M.B.*	7.72	3.89	10.84	13.9	7.16	17
Control	7.83	3.72	19.50	26.9	7.45	24

* M.B.= methyl bromide

Analyses performed at the Soil and Water Testing Laboratory, New Mexico State University

A. SEEDLING PRODUCTION (Cont.)

(3) Expt. 3: Nursery Production of Aspen on Neutral to Alkaline Nursery Soil.

Objective: Determine minimal soil physical amendments for aspen production in northern New Mexico.

This experiment was reported in the following paper presented at the 1983, Intermountain Nurseryman's Association Meeting.

Fisher, J.T. and G.A. Fancher. 1983. Effects of soil amendments on aspen seedling production, p. 66-68. In "The Challenge of Producing Native Plants for the Intermountain Area". Proc. Intermountain Nurseryman's Assoc. Conf., Aug. 8-11, 1983, Las Vegas, Nev.

EFFECTS OF SOIL AMENDMENTS ON ASPEN SEEDLING PRODUCTION

James T. Fisher and Gregory A. Fancher

ABSTRACT: Quaking aspen (*Populus tremuloides* Michx.) seedlings were grown in north central New Mexico in a mountain valley nursery soil amended with sulphur and one of four levels of peat moss (0, 1/4, 1/2 and 3/4 peat (v/v). The 1/4 peat treatment is equivalent to 374 m^3/ha. Peat moss improved soil medium physical and chemical properties responsible for improving seedling growth with each addition. Sulphur alone did not produce satifactory seedlings. Peat-amended soil produced plantable seedlings in one growing season at the study site.

INTRODUCTION

The geographical range of quaking aspen (*Populus tremuloides* Michx.) is enormous in western North America; it spans over 40° latitude. More than 200,00 hectares are occupied in New Mexico, Arizona and the adjacent San Juan Basin (Jones and Trujillo, 1975) where aspen forests provide numerous human benefits and renewable resources.

High on the list of potential benefits is the role aspen can play in redirecting the course of wildfire. In the southern Rockies, aspen has a lower fire potential than conifer types and can provide a critical fuelbreak. Flammability of aspen has been estimated to be less than one half that in adjacent conifers (Fechner and Barrows, 1976). This might explain why wildfires spreading from high elevation conifer forests have been observed to die out in aspen. Healthy stands of aspen are regarded by fire managers as relatively fire proof. It follows that maintenance and establishment of aspen are useful fire management practices, particularly in mountain resort areas where ignition is likely and the potential for loss of resource value and life is great.

At present, land managers in the Southwest do not possess a full understanding of the steps necessary to grow aspen seedlings reliably and efficiently, nor of those steps leading to fuelbreak establishment. Through a U.S. Forest Service-Eisenhower Consortium cooperative research project begun in 1981, we are developing or refining greenhouse, nursery, site preparation and weed control practices leading to establishment of aspen. This paper addresses bareroot seedling production.

Associate Professor of Woody Plant Physiology and Forest Research Specialist, respectively, Department of Horticulture, New Mexico State University, Las Cruces, N.M. 88003

Production of aspen seedlings from seed has been largely ignored in the West until recent years. However, large-scale production was achieved more than one decade ago in the Great Lakes region, notably at the Institute of Paper Chemistry (IPC), Appleton, Wisconsin (Benson and Dubey, 1972). The methods developed by IPC supplanted conventional nursery practices which generally failed to avoid:

(1) rapid loss of seed viability in the seedbed
(2) washing of the seed
(3) drying of the surface soil during the first two weeks
(4) damping-off during the seedling stage

The specific objective of this study was to apply IPC methods at a northern New Mexico mountain valley nursery site while testing soil amendments potentially useful in reducing soil pH and density. This refinement was believed necessary to avoid seedling disease and nutritional disorders, and to minimize nursery lifting difficulties.

METHODS AND MATERIALS

Site Description

The study was conducted at Mora Research Center located in north central New Mexico at an elevation of 2213 m. The frost free season is 100 to 120 days. Mean annual temperature is 6°C and mean annual precipitation is about 51 cm.

The study site is a level valley bottom. Soil is well drained alluvium with moderate to slow permeability. The upper 50 cm is a dark grayish brown (10YR 4/2) sandy clay loam. According to Cryer (1980) the soil profile classification is Cumulic Haploboroll.

Aspen seed used in this study was collected in early June, 1981 from open-pollinated clones growing from 2500 to 2700 m elevation about 15 km northeast of Santa Fe, New Mexico. At the time catkins were collected, seed release was just beginning on a few branches of sampled trees. Catkins were kept cool (18°C) during and following transfer to a laboratory and "cotton" was released and collected with a vacuum after 20 days. Harder's (1970) extraction procedure was used to remove "cotton" and minute debris. Cottony hairs of the placenta remaining attached to seeds can adversely affect germination removed (Myers and Fechner, 1980). Seed was bulked and stored at -4°C over anhydrous calcium sulfate ("Drierite") in a sealed jar to maintain

seed viability (Benson and Harder, 1972). Seed germination was above 90% when tested two weeks prior to nurserybed sowing.

Installation of experimental nursery beds followed procedures developed by Benson and Einsphar (1962) and modified by Benson and Dubey (1972). Within a 2.44 m x 15.9 m area, five 1.19 m x 2.41 m areas were excavated to a depth of 92 cm for each to accomodate a 1.22 m x 2.44m x 2.44 m wood frame supporting a hinged frame covered with standard window screen. Plywood boards divided each frame into equal quadrants to a depth of 92 cm. Polyethylene plastic lined the main frame soil side walls to the same depth.

The excavated soil was combined with horticulture-grade peat moss to establish four nursery bed growing media: (1) soil; (2) 1/4 peat, 3/4 soil; (3) 1/2 peat, 1/2 soil; and (4) 3/4 peat, 1/4 soil (by volume). In addition, elemental sulfur was added at the rate of 852 kg/ha (750 lb/ac) to each treatment. Physical and chemical properties of media were determined by routine soil test procedures employed by the Soil and Water Testing Laboratory, New Mexico State University.

Each bed frame was covered with plastic to fumigate all experimental plots with methyl bromide. The following day, frame tops were lifted and the beds were aerated for 48 hours.

Aspen seeds were sown at the spacing recommended by IPC (Benson and Dubey, 1972) to produce 110-150 seedlings per m^2. Following emergence, excess seedlings were thinned. Beds were irrigated daily by 1.8 cm bi-wall perforated drip tubing. Fertilizer was applied via irrigation water at the rate of 113 kg/ha N, 45 kg/ha P and 79.5 kg/ha K.

Treatments were randomized within frames. Within a 30 cm x 91 cm area centered within each quadrant, 12 seedings were labeled in order to record leaf number and height measurements, repeated at two-week intervals. Seedling density for each of three 30 cm x 30 cm subplots was recorded just prior to harvest.

Seventeen weeks from sowing, seedlings were lifted with a spade and enclosed in plastic bags. Ten trees were harvested from each subplot. Height, caliper, and fresh and oven dry weights were recorded for each seedling. A portable leaf area meter (Li-Cor, Inc.) was used to determine leaf area for 12 of the 30 seedlings harvested from each treatment. Analysis of variance, Duncan's mean separation test, and multiple linear regression were employed in data analyses.

RESULTS

Peat additions progressively improved physical and chemical properties of nursery bed media (Table 1). Most notable are improvements in soil reaction, pore space, hydraulic conductivity and cation exchange capacity. Organic matter increased considerably but approached the recommended level (3%) prior to any addition. In the field, soil peat moss reduced surface crusting and puddling compaction caused by irrigation.

Table 1. Chemical and Physical Properties of Nursery Bed Media

	SOIL	1/4 PEAT (v/v)	1/2 PEAT	3/4 PEAT
Hydraulic Conductivity (ml/cm^2 - hr)	14.6	30.6	93.3	245.2
Bulk Density (g/cc)	1.23	1.07	0.79	0.44
Pore Space (% By Vol.)	50.8	56.1	68.4	82.4
pH	7.4	6.8	6.0	4.8
% Organic Matter	2.5	4.0	7.9	15.6
C.E.C. (meq/100g)	14.1	15.5	21.0	39.0
Salts (% Sol.)	1.0	1.5	0.9	0.8
N-Total (PPM) (Kjeldahl)	894	1075	1160	2195
NO_3 (PPM)	13.5	22.6	29.9	42.9
P(PPM)	4.4	4.4	5.0	7.6
K(PPM)	11.6	18.5	19.6	29.8

* Before Addition of Sulfur.

Growth of seedlings grown with peat amendments were considerably taller and supported more leaves than those grown in soil alone (Figs 1-2). Seedling density averaged 132 per square meter across all treatments and density differences among treatments were not statistically significant at the .05 level. Table 2 compares harvested seedlings across treatments. Most significant is the failure of soil or soil and 1/4 peat to produce a minimum caliper of 0.3 cm (1/8"). Only 3/4 peat produced a 30-cm shoot. Reading across treatments in Table 2, differences for any paired numbers are statistically significant at the .01 level except leaf areas for 1/2 and 3/4 peat.
Multiple regression analysis of the pooled data provided an opportunity for examining growth

Figure 1. Cumulative Height Growth for Quaking Aspen Seedlings Under Nursery Bed Conditions

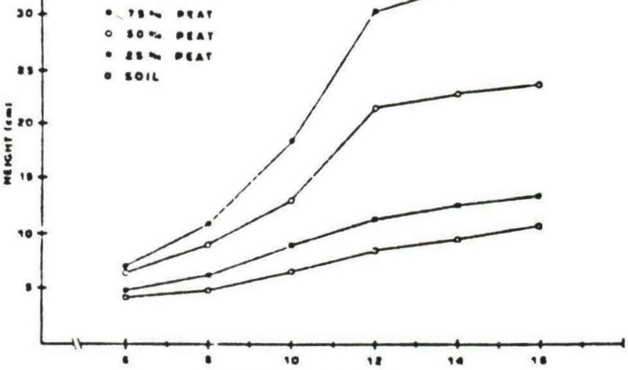


Figure 2. Cumulative Leaf Number for Quaking Aspen Seedlings Under Nursery bed conditions

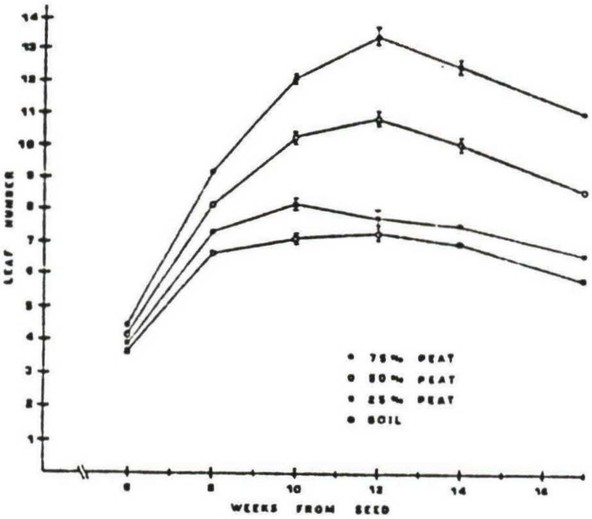


Table 2. Seedling Growth Responses at 16 Weeks

	SOIL	¼ PEAT	½ PEAT	¾ PEAT
Height (cm)	10.92	13.60	24.11	33.73
Caliper (mm)	1.94	2.26	3.18	3.95
Leaf Number	5.77	6.73	8.52	11.00
Leaf Area (cm^2)	21.88	30.29	49.32	50.16
Shoot DWT (g)	0.24	0.37	0.98	1.88
Root DWT (g)	0.11	0.22	0.57	0.99

relations of aspen seedlings. The correlation matrix found in Table 3 shows several parameters to be closely related. Specifically, height is closely related to caliper, leaf number, and shoot weight. All of the values shown are statistically significant (.0001 level).

Table 3. Correlation Matrix (R^2)

	Height	Caliper	Leaf No.	Shoot DWT	Root DWT	Leaf Area
Height	--	.86	.74	.81	.67	.22
Caliper		--	.68	.76	.71	.25
Leaf No.			--	.63	.52	.23
Shoot DWT				--	.78	.12
Root DWT					--	.12
Leaf Area						--

DISCUSSION AND CONCLUSIONS

The study demonstrated that plantable aspen seedlings can be successfully grown at the Mora Valley nursery site if the soil is amended with peat and sulphur. If the desired caliper is 0.3 to 0.9 cm (1/8" to 3/8"), 1/2 to 3/4 of the nursery medium must be peat if seedlings are grown and harvested in less than 110 days. In the Mora Valley, it would be possible to plant earlier, however, and this would result in larger seedlings. Allowed an additional three weeks, seedlings grown in 1/2 peat may reach desired dimensions.

The relative importance of physical and chemical conditions derived from peat were not determined. However, seedlings grown in peat-amended media were subjected to conditions more favorable than soil for nutrient exchange and uptake, and less favorable for build up of soil pathogens.

Applied over an extensive area, peat amendments would be costly and a local substitute might be sought. In northern New Mexico old composted sawdust can be obtained and may provide a satisfactory substitute (Montano, Fisher, and Cotter 1977). The disadvantages of fresh sawdust and farm yard manure were discussed by Armson and Sadreika (1974), who also recommended peat application rates and procedures.

LITERATURE CITED

Armson, K. A. and V. Sadreika. 1974. Forest tree nursery soil management and related practices. Can. Ministry of Natural Resources. 177 p.

Benson, M. K. and D. Dubey. 1972. Aspen seedling production in a commercial nursery. Inst. Pap. Chem. Genet. and Physiol. Notes No. 12, 7 p.

Benson, M. K. and Einsphar. 1962. Improved method for nursery production of quaking aspen seedlings. Tree Planters' Notes No. 53:11-14.

Benson, M. K. and M. L. Harder. 1972. Storage of aspen seed. Inst. Pap. Chem. Genet. and Physiol. Notes No. 11,4 p.

Cryer, D. H. 1980. Soil analysis: A method to determine Christmas tree productivity in the mountain valleys of Mora County. M.S. Thesis, New Mexico State Univ., Las Cruces. 110 p.

Harder, M. L. 1970. Procedures for collection and extraction of Populus seed. Inst. Paper Chem. Genet. and Physiol. Notes No. 9:3 p.

Jones, J. R. and D. P. Trujillo. 1975. Development of some young aspen stands in Arizona. USDA For. Serv. Res. Pap. RM-151,11 p.

Montano, J. M. J. T. Fisher and D. J. Cotter. 1977. Sawdust for growing containerized forest tree seedlings. Tree Planters' Notes 28:6-9.

Myers, J. F. and G. H. Fechner. 1980. Seed hairs and seed germination in Populus. Tree Planters' Notes 31:3-4.

B. ESTABLISHMENT OF ASPEN

(1) Expt. 5: Site preparation and Weed Control for Establishment of Aspen.

Objective: Develop reliable site prepartation and weed control practices for aspen forestation.

This experiment will be published as a journal article in the January 1987 issue of the Canadian Journal of Forest Science.

Fisher, J.T. and R.W. Neumann. 1987. Site preparation and weed control for aspen seedling establishment in the southeren Rocky Mountains. Can. J. For. Res. (in press).

Results were also presented at two conferences:

Fisher, J.T. and R.W. Neumann. Cultivation and herbicides promote quaking aspen seedling establishment. In "Weed Control for Forest Productivity". Spokane, Washington, Feb. 5-7, 1985 (in press).

Fisher, J.T. and R.W. Neumann. 1984. Western aspen seedling establishment: Site preparation. In Proc. 8th N. Amer. For. Biol. Wkshp, July 30-Aug. 1, 1984, Logan, Utah. (abstract).

SITE PREPARATION AND WEED CONTROL FOR ASPEN SEEDLING ESTABLISHMENT IN THE SOUTHERN ROCKY MOUNTAINS[1]

James T. Fisher and Robert W. Neumann

Department of Horticulture, New Mexico State University

Las Cruces, New Mexico, U.S.A. 88003

Phone: (505) 646-1521

---

[1]New Mexico Agric. Exp. Sta. journal article no. 1241.

ABSTRACT

Fisher, J. T., and R. W. Neumann. 1986. Site preparation and weed control for aspen seedling establishment in the southern Rocky Mountains. Can. J. For. Res.

Containerized aspen (Populus tremuloides Michx.) seedlings were planted at high elevation sites in southern (May 1982) and northern (July 1983) New Mexico. Each plantation compared fall cultivation (20 cm depth), prior to planting the following spring or summer, with cultivation at the time of planting. Subtreatments of the tests included applications of the postemergent herbicide dalapon and the preemergents linuron, trifluralin or simazine.

First season survival exceeded 75% for the best treatment at each site. Cultivation, in general, effectively reduced weed cover and improved seedling success. Fall cultivation, in particular, improved seedling survival and growth only at the relatively dry southern site. Except for spring-cultivated plots in the south, some herbicide applications improved weed control and seedling performance over cultivation alone. The combination of fall cultivation plus trifluralin is considered the best site preparatory treatment tested.

INTRODUCTION

The geographic range of quaking aspen (Populus tremuloides Michx.) in western North America spans more than 40 degrees latitude. More than 200,000 hectares are occupied in New Mexico, Arizona and the adjacent San Juan Basin (Jones and Trujillo 1975) where aspen forests provide many human benefits and renewable resources.

An important potential benefit of aspen is its role in redirecting the course of wildfire. In the southern Rockies, aspen has a lower fire potential than conifer types, and can provide a critical fuelbreak. Aspen flammability has been estimated to be less than half that of adjacent conifers (Fechner and Barrows 1976). This might explain why wildfires spreading from high elevation conifer forests have been observed to die out in aspen. Healthy aspen stands are regarded by fire managers as relatively fireproof. It follows that maintenance and establishment of aspen are useful fire management practices, particularly in mountain resort areas where ignition is likely and the potential for loss of resource value and life is great.

At present, land managers in the Southwest do not have a full understanding of the steps necessary to grow aspen seedlings reliably and efficiently, or of those steps leading to fuelbreak establishment. Through a U.S. Forest Service-Eisenhower

Consortium cooperative research project begun in 1981, we are developing or refining greenhouse, nursery, site preparation and weed control practices leading to aspen establishment. This paper addresses the last two elements of the project, site preparation and weed control.

METHODS

Two separate experiments were established, one in the Sacramento Mountains of south-central New Mexico in May 1982, the other 14 months later in the Sangre de Cristo Mountains, 360 kilometers to the north. The southern site (elevation 2,650 m) annually receives approximately 700 mm of precipitation, 550 mm of which are lost to evapotranspiration. The northern site (elevation 2,870 m) is more moist, averaging 900 mm precipitation and 400 mm evapotranspiration. Plantations were not irrigated.

Similar split-block experimental designs were used for both studies. At each site, main treatments tested time of pre-plant cultivation:

1. fall cultivation (after treatment with dalapon[2] at 5.6 kg ai/ha)

2. spring (south) or summer (north) cultivation

Subtreatments at each site tested herbicides and cultivation:

1. linuron[3] at 1.1 kg ai/ha
2. linuron at 2.2 kg ai/ha
3. trifluralin[4] at 1.1 kg ai/ha
4. trifluralin at 2.2 kg ai/ha
5. simazine[5] at 2.7 kg ai/ha

10 6. simazine at 4.5 kg ai/ha

11 7. cultivation alone (no chemical applications)

12 8. dalapon at 5.6 kg ai/ha before cultivation (fall only)

13 9. untreated (spring or summer only)

---

[2] 2,2-dichloropropanoic acid

[3] *N*'-(3,4-dichlorophenyl)-*N*-methoxy-*N*-methylurea

[4] 2,6-dinitro-*N*,*N*-dipropyl-4-(trifluoromethyl)benzenamine

[5] 6-chloro-*N*,N'-diethyl-1,3,5-triazine-2,4-diamine

In September of the year before planting, the postemergent herbicide dalapon was applied to fall main plots at both sites, excluding the cultivation alone subplot. When the vegetation browned about 2 weeks later, plots were cultivated to a depth of approximately 20 cm. The dalapon subtreatment received no additional herbicides. Spring cultivation and application of preemergent herbicides linuron, trifluralin and simazine was completed the following April at the southern site. The same was done in early July at the northern site. All chemicals were applied with a calibrated, hand-held boom sprayer. Trifluralin was incorporated into the soil according to label directions.

Four-month-old containerized seedlings were sorted by size to ensure uniformity within each replication, and were planted in May 1982 in the south and July 1983 in the north (2 to 3 weeks after the herbicide applications at each site). Seedling containers were 160 cm^3 (south) and 350 cm^3 (north). Seedlings planted at the southern site were pruned to a uniform height of 20 cm after planting. Forty-tree rectangular plots were replicated four times at each site.

Seedling survival was evaluated 4 months after planting in the south and 11 months after planting in the north. Percent soil moisture was determined at planting and regularly for several

weeks thereafter. Weed cover was evaluated periodically after planting by the point sampling method (Grieg-Smith 1957).

Height growth was measured at the southern site 1 year after planting. Height and basal stem caliper were measured 14 months after planting the northern site.

Data were were analyzed as plot means via the protected LSD. Statistical differences discussed in this article are at the $p \leq .05$ level.

RESULTS

The Southern Site (Table 1)

SURVIVAL--Seedling survival declined during the first growing season as mid-summer drought took its toll on many seedlings in the study. By September 1982, nearly 100% mortality in untreated plots showed that cultivation, regardless of timing, markedly improved seedling survival. Survival for fall-cultivated plots was 49.2%, and was statistically greater than 40.4% survival in spring-cultivated plots. Soil moisture at planting was significantly greater for fall-cultivated plots than it was for those cultivated in spring. Main treatment differences for soil moisture diminished as early-season rains arrived, but fall cultivation, and its associated winter fallowing, resulted in

additional soil moisture during the critical establishment period, and effectively increased seedling survival.

Fall/dalapon resulted in higher seedling survival (75.6%) than did other fall subtreatments, except trifluralin (63.5% for the high rate, 56.5% for the low rate) and linuron at the low rate (61.0%). Excluding the untreated control, survival rates for spring subtreatments did not differ significantly.

In both fall- and spring-cultivated plots, survival for cultivation alone plots was equal to or greater than survival for preemergent herbicide plots. That is, preemergent herbicides did not improve seedling survival in cultivated plots.

HEIGHT--Plantation-wide height was reduced 60-70% by first season main stem dieback. In May 1983, seedlings in fall plots averaged 9.1 cm in height and were significantly taller than seedlings in spring plots (6.6 cm). The only cultivation/herbicide combination to result in superior growth was fall/trifluralin (2.2 kg ai/ha), which produced seedlings averaging 12.0 cm tall.

WEED CONTROL--Perennial grass sod, composed primarily of western wheatgrass (*Agropyron smithii* Rudb.) and Kentucky bluegrass (*Poa pratensis* L.), covered more than 75% of the ground area before cultivation. Western yarrow (*Achillea lanulosa*

Nutt.), dandelion (*Taraxacum officinale* Weber), vetch (*Vicia sativa* L.) and other forbes were present among the grasses.

Two months after the May planting, percent weed cover did not differ significantly between cultivation times. Fall and spring plots averaged 40.8 and 42.2% cover, respectively. Preemergent herbicides, associated with either cultivation time, did not differ in their respective effects on weed density. Among fall-cultivated subplots, only simazine at 2.7 kg ai/ha (27.8% cover), linuron at 2.2 kg ai/ha (35.0% cover) and trifluralin at 2.2 kg ai/ha (38.3% cover) provided significantly better weed control than cultivation alone (57.5% cover). Spring/cultivation alone (43.8% cover) controlled weeds as well as any spring herbicide subtreatment, but all spring-cultivated subplots were significantly less weedy than the untreated control (76.5% cover).

By July 1983, overall weed cover had risen to 47.0% for fall-cultivated plots and to 50.8% for plots cultivated in spring, but differences remained insignificant. Control (untreated) plots averaged 61.8% cover, but no statistical differences were detected among subtreatments within either cultivation time. Composition of the weed species did not change over the test period.

The Northern Site (Table 2)

SURVIVAL--After experiencing generally poor survival at the southern site, northern site seedlings were grown in larger containers (350 cm^3) and planted at a later date (July) to avoid mid-summer drought. Plantation-wide survival was high, but because of the late planting, winter fallowing effects associated with fall cultivation were diminished after the arrival of early spring rains. However, moisture at a soil depth of 15 to 30 cm was still significantly greater at planting for fall-cultivated plots than for those cultivated in summer.

Eleven months after planting, survival for main treatments was 70.9% for fall plots and 67.1% for summer, but the difference was not significant. Within both fall and summer plots, no preemergent herbicide treatment produced a survival rate superior to that for cultivation alone.

HEIGHT--Initial seedling heights averaged 32 cm, ranging from 19 to 47 cm. Seedling dieback was not observed over the course of the study. Seedlings from summer plots averaged 51.9 cm tall and were not significantly taller than seedlings in fall plots (47.9 cm). Among fall subtreatments, the two trifluralin treatments resulted in significantly taller seedlings (55.3 cm for the low rate, 53.8 cm for the high rate). Seedlings in summer/trifluralin (2.2 kg ai/ha) plots averaged 57.6 cm and were significantly

taller than those in other summer subtreatments, except those in plots treated with trifluralin at 1.1 kg ai/ha (55.5 cm) or simazine at 2.7 kg ai/ha (53.1 cm). Among summer subtreatments, seedling height was least in untreated plots (31.5 cm).

BASAL STEM CALIPER--At planting, seedlings averaged 2.2 mm basal stem caliper, ranging from 1.5 to 3.0 mm. Fourteen months later, summer plot seedlings averaged 5.9 mm basal stem caliper and were statistically equivalent to those from fall plots (5.6 mm). As with height, stem caliper was greatest in fall/trifluralin plots, averaging 6.3 and 6.2 mm in basal stem caliper for the low and high rates of the herbicide, respectively. Among summer subtreatments, trees in trifluralin at 2.2 kg ai/ha plots (6.3 mm) had stem calipers larger than those in plots treated with linuron at 1.1 kg ai/ha (5.7 mm), simazine at 2.7 kg ai/ha (5.6 mm), cultivation alone (5.8 mm) or untreated (3.8 mm). The untreated plot produced seedlings with smallest stem caliper.

WEED CONTROL--At the northern site, a dense sod occupied more than 90% of the area within plots before cultivation. Weeds included Kentucky bluegrass, timothy (Phleum pratense L.), western yarrow, dandelion, iris (Iris missouriensis Nutt.) and others.

Four months after planting, weed cover averaged 48.2% in summer- and 57.8% in fall-cultivated plots, but the difference was not significant. The low rate of trifluralin with fall

cultivation provided better control (30.5% cover) than other fall treatments, except trifluralin at 2.2 kg ai/ha (33.8% cover) and linuron at 2.2 kg ai/ha (52.5% cover). Summer cultivation/herbicide subtreatments did not differ, but applications of trifluralin at both rates (35.3% cover for the low rate, 38.0% for the high rate) or simazine at the high rate (36.0% cover) provided better weed control than summer cultivation alone. Weed cover exceeded 95% in untreated control plots.

By September 1984, percent weed cover had increased considerably over what it was 10 months earlier and, again, cultivation times did not differ significantly (77.8% cover for fall plots, 79.4% cover for summer). Trifluralin (2.2 kg ai/ha) was the top-ranked fall subtreatment (63.3% cover), statistically equalled only by trifluralin (1.1 kg ai/ha) with 69.8% average weed cover. Cultivation alone (81.5% cover) controlled weeds as effectively as any herbicide tested with summer cultivation. Weed cover for the untreated plot averaged 94.3% and did not differ significantly from the 90.5% of the summer/linuron (1.1 kg ai/ha) plot. Composition of the weed species did not change over the test period.

DISCUSSION AND CONCLUSIONS

Dense, perennial sod poses a formidable threat to aspen seedling establishment. Sod cultivation simplifies planting and reduces weed competition. Cultivation was the single most effective treatment tested, evidenced by increased seedling survival and growth in cultivated plots. At the southern site, seedling survival was significantly greater in cultivated plots than in untilled plots. In the north, cultivation did not influence survival, but did improve growth.

At the southern site, seedlings planted in fall-cultivated plots apparently benefitted from additional soil moisture stored during the fallow condition. At the northern site, moisture captured after fall cultivation improved growth, but apparently was not required for survival.

Preemergent herbicide applications did not benefit seedling survival at either site, and fall cultivatioin plus simazine at the high rate was phytotoxic to seedlings. However, trifluralin effectively and persistently reduced weed competition without damaging aspen seedlings in northern fall plots, and seedlings responded with superior growth. At the southern site, seedlings grew taller in fall/trifluralin (2.2 kg ai/ha), although weed competition was equal to that in other plots.

Because moisture stress and weeds reduced either seedling growth or survival, or both, aspen planting sites should be cultivated. The moisture conserved through fall cultivation improves aspen seedling establishment, depending on the scarcity of soil moisture. Results from the southern site indicate dalapon applied before fall cultivation improves seedling survival over cultivation alone. Preemergent herbicide applications will not benefit seedling survival, but can improve seedling growth through better weed control. The fall cultivation/trifluralin treatment is considered the most effective site preparatory treatment tested.

ACKNOWLEDGEMENTS

We wish to express our appreciation to the Eisenhower Consortium and the Forest Service--USDA for funding, through grant number RM-81-160-GR, and to the Lincoln and Santa Fe National Forests for making land available for study sites. We are also grateful to John B. McRae for the dedication and long hours he contributed to the project, and to Dr. Winston Richards and the Department of Experimental Statistics at New Mexico State University for analytical support.

LITERATURE CITED

Fechner, G. H. and J. S. Barrows. 1976. Aspen stands as wildfire fuel breaks. Eisenhower Consortium Bull. 4, 26 p.

Grieg-Smith, P. 1957. Quantitative Plant Ecology. Butterworths. London. 198 p.

Jones, J. R. and D. P. Trujillo. 1975. Development of some young aspen stands in Arizona. USDA For. Serv. Res. Pap. RM-151, 11 p.

Little, T. M. and F. J. Hills. 1978. Agricultural Experimentation Design and Analysis. John Wiley and Sons. New York. 350 p.

Table 1. Results for the southern site (planted May 1982).

Treatment	Soil Moisture (%) at Planting 0-15cm depth	Soil Moisture (%) at Planting 15-30cm depth	Survival (%) Sep 82	Height (cm) May 83	Weed Cover (%) Jul 82	Weed Cover (%) Jul 83
Cultivation Times*						
Fall	23.7 a	30.7 a	49.2 a	9.1 a	40.8 a	47.0 a
Spring	17.1 b	24.8 b	40.4 b	6.6 b	42.2 a	50.8 a
Fall Cultivation Subtreatments						
linuron (1.1)			61.0 ab	7.4 b	43.3 abc	42.8 a
linuron (2.2)			51.9 b	9.0 b	35.0 a	56.5 a
trifluralin (1.1)			56.5 ab	8.8 b	42.0 abc	45.8 a
trifluralin (2.2)			63.5 ab	12.0 a	38.3 ab	41.8 a
simazine (2.7)			44.8 b	9.2 b	27.8 a	46.8 a
simazine (4.5)			22.1 c	7.0 b	41.5 abc	41.3 a
cultivation alone			44.8 b	8.6 b	57.5 c	54.3 a
dalapon			75.6 a	8.4 b	52.8 bc	47.0 a
Spring Cultivation Subtreatments						
linuron (1.1)			42.3 a	7.1 a	46.3 a	54.0 a
linuron (2.2)			46.5 a	6.3 a	41.8 a	58.0 a
trifluralin (1.1)			44.2 a	5.7 a	47.8 a	55.0 a
trifluralin (2.2)			37.5 a	6.1 a	32.3 a	44.8 a
simazine (2.7)			39.8 a	7.0 a	43.8 a	52.3 a
simazine (4.5)			29.2 a	6.5 a	40.0 a	53.5 a
cultivation alone			43.8 a	7.0 a	43.8 a	38.3 a
untreated			0.6 b	8.3 a	76.5 b	61.8 a

Note: Column values with the same letter are not significantly different ($p \leq .05$) as determined by the protected LSD. Survival and weed cover data were subjected to arcsine transformation (Little and Hills 1978) prior to analysis. Means presented are for the untransformed data.

*The seven subtreatments common to both main treatments (cultivation times) are combined for analysis.

Table 2. Results for the northern site (planted July 1983).

Treatment	Soil Moisture (%) at Planting 0-15cm depth	Soil Moisture (%) at Planting 15-30cm depth	Survival (%) Jun 84	Height (cm) Sep 84	Caliper (mm) Sep 84	Weed Cover (%) Nov 83	Weed Cover (%) Sep 84
			Cultivation Times*				
Fall	31.1 a	31.9 a	67.1 a	47.9 a	5.6 a	57.8 a	77.8 a
Summer	26.9 a	23.7 b	70.9 a	51.9 a	5.9 a	48.2 a	79.4 a
			Fall Cultivation Subtreatments				
linuron (1.1)			69.4 ab	47.6 b	5.2 bc	74.5 cde	89.0 d
linuron (2.2)			78.1 a	48.2 b	5.6 b	52.5 abc	79.5 cd
trifluralin (1.1)			68.8 ab	55.3 a	6.3 a	30.5 a	69.8 ab
trifluralin (2.2)			73.1 a	53.8 a	6.2 a	33.8 ab	63.3 a
simazine (2.7)			66.3 abc	43.7 bc	5.3 bc	74.3 de	81.5 cd
simazine (4.5)			43.8 c	47.9 b	5.5 b	55.5 bcd	77.0 bc
cultivation alone			70.0 ab	40.2 cd	4.8 cd	83.3 e	84.8 cd
dalapon			50.0 bc	35.1 d	4.5 d	88.3 e	87.8 d
			Summer Cultivation Subtreatments				
linuron (1.1)			73.1 ab	49.1 cd	5.7 bc	52.0 ab	90.5 b
linuron (2.2)			78.1 a	51.2 bcd	5.9 abc	54.5 ab	80.8 a
trifluralin (1.1)			78.1 a	55.5 ab	6.2 ab	35.3 a	76.3 a
trifluralin (2.2)			71.3 ab	57.6 a	6.3 a	38.0 a	71.5 a
simazine (2.7)			55.6 b	53.1 abc	5.6 c	53.8 ab	81.8 a
simazine (4.5)			66.9 ab	47.4 d	5.8 abc	36.0 a	74.8 a
cultivation alone			73.1 ab	49.1 cd	5.6 c	67.8 b	80.5 a
untreated			53.8 b	31.5 e	3.8 d	95.3 c	94.3 b

Note: Column values with the same letter are not significantly different ($p \leq .05$) as determined by the protected LSD. Survival and weed cover data were subjected to arcsine transformation (Little and Hills 1978) prior to analysis. Means presented are for the untransformed data.

*The seven subtreatments common to both main treatments (cultivation times) are combined for analysis.

A. SEEDLING PRODUCTION (Cont.)

(4). Exp. 4. Modification of containerized aspen roots

Objective: Determine container seedling response to $CuCO_3$ applied as a chemical root pruning treatment

ASPEN CONTAINER SEEDLING RESPONSE TO CHEMICAL ROOT PRUNING TREATMENTS

Ryder, J., J.T Fisher and J.G. Mexal

Department of Agronomy and Horticulture
New Mexico State University
Las Cruces, NM 88003

INTRODUCTION

Tree seedlings commonly have malformed root systems when grown in containers, particularly when containers lack inner wall ribs to prevent root spiralling. Coiled, strangulated roots severely can restrict growth, or fail to anchor young trees. Studies have shown that conifer seedling root morphology can sometimes be controlled by the judicious use of root pruning chemicals and appropriate container volume. Root pruning chemicals have reduced greatly the occurrence and severity of conifer root malformation (Burdett, 1978; Pellett *et al*., 1980; McDonald *et al*., 1984a, Ruehle 1985). Because growth is arrested when main lateral roots contact the pruning compound, roots do not deflect down the container wall or form a contorted root mass. Root pruning often enhances the development of conifer feeder roots and subsequent seedling growth, depending on species. Because seedling size is directly related to container volume, rooting volumes poorly matched with production schedules result in stunted, root-bound seedings (Endean and Carlson, 1975). Container volume influences transplant survival, evidenced by southern pine seedling survival data reported by Amidon *et al*., 1981.

Hardwood seedling response to chemical pruning has received minor attention in the literature. This study examined the effects of container volume, cupric carbonate and latex paint treatments on the growth and development of *Populus tremuloides* container seedlings.

MATERIALS AND METHODS

The study compared aspen seedling response to five container wall treatments and two book planter container volumes (165 and 350 cm^3). Cupric carbonate ($CuCO_3$)

was mixed with exterior Pittsburgh acrylic latex flat paint and applied to inner walls of open book planters at levels of 0, 25, 50 and 100 g of $CuCO_3$/ liter of paint. A control treatment with paint only tested the effect of the paint in the absence of $CuCO_3$.

Seed Collection

Aspen catkins were collected in June 1985 from trees growing near Lake City, Colorado (Gunnison National Forest). Seeds were extracted and stored in accordance with recommended procedures (Harder 1970, Benson and Harder 1972). Seeds were sown in 3-cm^3 plug cavities at a density of 2400 plugs/ m^2. Three weeks after sowing, the 5-cm tall plug seedlings were transplanted into book planters. The book planter growing medium was a 1:1 (v/v) mix of moist peat and perlite.

Seedlings in plug trays were irrigated daily with Hoagland's complete nutrient solution. Book planters were irrigated with Hoagland's on alternate days. The solution contained the following nutrients mixed in 10 liters of water:

Fe chelate--------23 g
H_3PO_4--------------37 ml
$MgSO_4$-------------176 g
KNO_3---------------60 g
Urea---------------62 g
NH_4NO_3------------305 g

This nutrient solution was applied through a proportioner at a 1:128 ratio. Greenhouse temperature was 20-25 C and photoperiod was artificially extended to 18 hours with incandescent lights

Seedling Measurements

Seedling height and root collar diameter were recorded bi-weekly, beginning one month after transplanting aspen plugs. Seedlings were harvested 10 and 22 weeks after transplanting to measure shoot height, root collar diameter, root and shoot oven dry weight, and leaf area as determined with a Li-Cor leaf area meter. For each harvest, ten trees were taken from each treatment and replicate. Root surface area was determined on a subsample of roots collected during the final harvest. Surface area of fresh roots were determined with a Li-Cor Leaf Area Meter. Root surface area was obtained from a regression equation previously determined (R^2=.96), relating Li-Cor area readings to actual root segment surface areas (Y=-6.96 + 2.51 X). Dry weights of main structural roots and fibrous roots were also determined to derive % fibrous roots.

Experimental Design and Data Analysis

The experimental design was a RCB with a five by two factorial treatment design. Twenty-four seedlings were grown for each treatment and each treatment was replicated three times. Bi-weekly measurements were taken on the same 10 seedlings randomly selected at the time of initial data collection in each treatment and replicate.

Data were analyzed using analysis of variance techniques; treatment means were separated with Duncan's Multiple Range test.

RESULTS

Container volume significantly affected seedling growth (Tables 1-3). Seedlings grown in the larger, 350-cm^3 container consistently had greater total biomass, shoot biomass and leaf biomass. Seedling height, root collar diameter and leaf area were consistently greater in the larger container only at the 10-week harvest (Table 3).

Most notable among results is the absence of any positive effects associated with pruning treatments on seedling root structure or growth (Tables 1-4). Specifically, chemical treatments did not significantly increase seedling root fibrosity above controls (Table 4), did not systematically alter R/S, and did not improve seedling growth.

Container volume and container wall treatment interaction was significant only for caliper measurements at the time of final harvest. Seedlings grown in the 350-cm^3 paint-only containers had calipers equal to calipers of seedlings grown in the 165-cm^3 containers, regardless of container wall treatment. Moreover, the paint-only treatment applied to the 350-cm^3 container consistently reduced seedling shoot, root and leaf biomass below control seedlings (Table 2).

DISCUSSION AND CONCLUSIONS

In a similar study (Fisher *et al*. 1986), $CuCO_3$ applied at 25 g/l, effectively pruned *Pinus caribaea* lateral roots in 41- and 165-cm^3 containers. However, the 50 or 100 g/l levels were required to produce similar results in 350- and 740-cm^3 containers. This trend was not detected in this study.

McDonald *et al*. (1984b) grew *P. ponderosa* and *P. contorta* in $CuCO_3$-treated containers and found effective pruning of primary laterals and subsequent proliferation of short roots. Similarly, $CuCO_3$ caused

Table 1. Aspen seedling biomass response to container volume and chemical root pruning treatments 10 weeks after transplanting.

Treatment	biomass (g dry wt.) total shoot	root	shoot	leaf	R/S
165 cm^3					
control	0.18 d	0.15 b	0.07 d	0.11 c	0.75 a
paint	0.12 e	0.08 c	0.04 f	0.08 c	0.58 cd
25 g/l	0.15 de	0.08 c	0.05 ef	0.10 c	0.51 cd
50 g/l	0.16 de	0.08 c	0.06 de	0.10 c	0.46 d
100 g/l	0.16 de	0.09 c	0.06 ef	0.10 c	0.61 bc
350 cm^3					
control	0.27 c	0.17 ab	0.10 c	0.17 b	0.66 ab
paint	0.31 bc	0.16 b	0.11 bc	0.20 ab	0.52 cd
25 g/l	0.36 bc	0.20 a	0.12 a	0.24 a	0.64 abc
50 g/l	0.30 bc	0.18 ab	0.11 abc	0.19 ab	0.64 abc
100 g/l	0.33 ab	0.19 a	0.12 ab	0.21 ab	0.59 bc

Means within columns are not significantly different (0.05 level) if followed by the same letters

Table 2. Aspen seedling biomass response to container volume and chemical root pruning treatments 22 weeks after transplanting.

Treatment	biomass (g dry wt.) total shoot	root	shoot	leaf	R/S
165 cm^3					
control	1.03 d	0.55 cde	0.51 d	0.52 de	0.59 ab
paint	0.91 d	0.39 e	0.40 d	0.51 e	0.42 c
25 g/l	1.06 d	0.43 de	0.51 d	0.55 de	0.48 abc
50 g/l	0.93 d	0.44 de	0.45 d	0.48 e	0.59 ab
100 g/l	1.11 d	0.54 cde	0.54 d	0.55 de	0.46 bc
350 cm^3					
control	2.05 ab	1.19 a	1.06 ab	0.98 ab	0.64 a
paint	1.34 cd	0.64 cd	0.64 cd	0.70 cd	0.47 bc
25 g/l	2.00 ab	0.97 b	1.02 ab	0.98 ab	0.52 abc
50 g/l	1.69 bc	0.85 bc	0.84 bc	0.85 bc	0.47 bc
100 g/l	2.24 a	1.22 a	1.13 a	1.11 a	0.55 abc

Means within columns are not significantly different (0.05 level) if followed by the same letters.

Table 3. Aspen seedling height, root collar diameter and leaf area response to container volume and chemical root pruning 10 and 22 weeks after transplanting.

	Ht. (cm)		Root collar dia. (mm)		Leaf Area (cm^2)	
	weeks					
	10	22	10	22	10	22
165 cm^3						
control	12.9 c	34.6 cd	1.7 c	3.0 cd	29.9 c	128.4 c
paint	9.8 e	28.4 d	1.6 c	2.8 d	21.7 d	108.8 c
25 g/l	10.9 de	33.3 cd	1.6 c	2.9 cd	24.8 cd	122.1 c
50 g/l	11.8 cd	30.3 d	1.6 c	2.9 d	27.4 cd	106.7 c
100 g/l	11.9 cd	38.9 bc	1.6 c	3.1 cd	26.0 cd	135.0 c
350 cm^3						
control	16.2 b	53.7 a	1.9 b	3.8 ab	43.7 b	225.6 ab
paint	15.6 b	32.4 cd	2.1 a	2.9 d	49.8 ab	139.7 c
25 g/l	17.5 a	46.2 ab	2.2 a	3.9 a	53.8 a	207.8 ab
50 g/l	15.9 b	45.4 ab	2.1 a	3.4 bc	49.4 ab	185.8 b
100 g/l	16.0 b	51.8 a	2.1 a	4.0 a	55.4 a	234.1 a

Means within columns are not significantly different (0.05 level) if followed by the same letters.

Table 4. Aspen root morphology response to container volume and chemical root pruning.

	Root Surface Area (cm^2) Total Fresh	% Fibrous (fibrous dry wt./ tot. root dry wt.) x 100
165 cm^3		
control	69.9 bcd	24
paint	35.0 d	19
25 g/l	52.0 d	20
50 g/l	39.4 d	16
100 g/l	52.3 d	18
350 cm^3		
control	124.3 a	28
paint	71.3 bcd	30
25 g/l	93.4 abc	17
50 g/l	100.8 ab	24
100 g/l	59.3 dc	26

Means within columns are not significantly different (0.05 level) if followed by the same letters.

proliferation of secondary and higher order laterals of *P. caribaea* (Fisher *et al.* 1986), but did not influence the number of primary lateral roots.

McDonald *et al.* (1984b) and Fisher *et al.* (1986) reported that $CuCO_3$ improved conifer seedling shoot growth, possibly because root pruning increased short root surface area. In contrast to these studies, $CuCO_3$ did not increase aspen seedling growth above controls, regardless of container size. In fact, the paint-only treatment generally reduced growth. Latex paint and $CuCO_3$ equally stimulated *P. caribaea* seedling growth, but the addition of $CuCO_3$ was necessary to prevent root deflection (Fisher *et al.* 1986).

Root hierarchy, evident in the pine seedling root system, was less obvious in the aspen seedling root system. The secondary, tertiary and higher order lateral (short) roots routinely observed in pine seedlings, and primarily responsible for nutrient absorption, were much less obvious in the aspen root. The aspen root system is essentially a structural matrix of the central tap root and primary laterals that bears a network of fibrous roots. Perhaps the absence of a strongly expressed heirarchical root system accounts for the lack of a positive pruning response to the container wall treatments, including stimulation of growth. The fibrous roots may be more fragile and therefore susceptible to Cu and/or paint toxicity (Table 4) than most *Pinus* short roots.

This study underscores the potential differences among conifer and hardwood species in their response to $CuCO_3$. Differences among conifers were previously noted: In an earlier study, Burdett and Martin (1982) found that the effectiveness of chemical root pruning treatments varied among several conifers and in one experiment was consistently ineffective with *Picea glauca*, *Picea sitchensis*, *Abies amabilis*, *P. contorta*, *Pseudotsuga menziesii*, *Thuja plicata* and *Tsuga heterophylla*.

Copper compounds have been applied to the bottom of flats to prune seedling roots of several ornamentals (Nussbaum 1969, as cited by Burdett and Martin 1982). The resultant root systems were compact and fibrous, and were suitable for tranplanting. Because the aspen root system is naturally fibrous, it was believed possible that the use of a chemical root pruner might increase fibrosity. However, in this study, chemical root pruning did not increase aspen root fibrosity, and stressed seedlings in some treatments resulting in decreased growth. Overall, we conclude that $CuCO_3$ does not significantly improve aspen seedling growth or quality when used as applied in this study. Plantable aspen seedlings grown in the 350-cm^3 untreated containers for 20-22 weeks were judged equal or superior to seedlings grown under $CuCO_3$ treatments.

LITERATURE CITED

Amidon, T.E., Barnett, J.P., Gallagher, H.P.and McGilvray, J.M. 1981. A field test of containerized seedlings under drought conditions, p. 139-145. In Proc. of the Southern Containerized Forest Tree Seedling Conf. Savannah, Ga., Aug. 25-26, 1981.

Benson, M.K. and M.L. Harder. 1972. Storage of aspen seed. Inst. Pap. Chem. Genet. and Physiol. Notes No. 11, 4 p.

Burdett, A.N. 1978. Control of root morphogenesis for improved mechanical stability in container-grown lodgepole pine. Can. J. For. Res. 8:483-486.

Burdett, A. N. and P. A. Martin. 1982. Chemical root pruning of coniferous seedlings. HortScience 17:622-624.

Carlson, L. and Endean, F. 1976. The effect of rooting volume and container configuration on the early growth of white spruce seedlings. Can. J. For. Res. 6:221-224.

Fisher, J.T., A. Romero and J. Ryder. 1986. Growth response of *Pinus caribaea* to cupric carbonate and container volume, p. 70-76. In Proc. 9th N. Amer. Fo. Biol. Wkshp, June 15-18, 1986, Okal. St. Univ., Stillwater, OK.

Harder, M.L. 1970. Procedures for collection and extraction of *Populus* seed. Inst. Pap. Chem. Genet. and Physiol. Notes No. 9, 3 p.

McDonald, S.E., Tinus, R.W., Reid, C.P.P. and Grossnickle, S.C. 1984a. Effect of $CuCO_3$ container wall treatment and mycorrhizae fungi inoculation of growing medium on pine seedling growth and root development. J. Environ. Hort. 2:5-8.

McDonald, S.E., Tinus, R.W. and Reid, C.P.P. 1984b. Modification of ponderosa pine root system in containers. J. Environ. Hort. 2:1-5.

Ruehle, J.L. 1985. The effect of cupric carbonate on root morphology of containerized mycorrhizal pine seedlings. Can. J. For. Res. 15:586-592.

(C) ASPEN SEEDLING STOCK OUTPLANTING SUCCESS

(1) Expt. 6: Suitability of Four Commercial Container Systems for Aspen Production and Outplanting.

Objective: Compare spring and summer outplanting survival of container and bareroot seedlings.

This experiment was recently presented at the Ninth North American Forest Biology Workshop. It will be published in 1986 as:

Fisher, J.T., R.W. Neumann and D.J. Manuchia. 1986. Suitability of aspen planting stock for establishment in the southern Rockies, p. 63-69. In Proc. 9th N. Amer. For. Biol. Wkshp, June 15-18, 1986, Oklahoma State University, Stillwater, OK.

A comparison of production costs for the stock types is presented in Appendix Table 1 which follows the paper cited above.

SUITABILITY OF ASPEN PLANTING STOCK FOR ESTABLISHMENT IN THE SOUTHERN ROCKIES

J.T. Fisher, R.W. Neumann and D.J. Manuchia[1]

Abstract.-- Four aspen (*Populus tremuloides* Michx.) seedling stock types were planted at two high elevation sites in southern (1982) and northern (1983) New Mexico. Each study independently evaluated early (May) and late (July) plantings, and stock types: bareroot and 164-cm^3, 350-cm^3 and 870-cm^3 containers. Site and planting date influenced stock type performance. July is the superior planting date for container seedlings. Survival was generally highest among the 164-cm^3 and 350-cm^3 stock types with July planting improving survival on the southern site and growth on the northern site.

Additional keywords: *Populus tremuloides*, hardwood stock type, fire fuel break, forestation.

INTRODUCTION

The geographic range of quaking aspen (*Populus tremuloides* Michx.) in western North America spans more than 40 degrees latitude. More than 200,000 hectares are occupied in New Mexico, Arizona and the adjacent San Juan Basin (Jones and Trujillo 1975) where aspen forests provide many human benefits and renewable resources.

An important benefit of aspen is its role in redirecting the course of wildfire. In the southern Rockies, aspen has a lower fire potential than conifer types, and can provide a critical fuelbreak. Aspen flammability has been estimated to be less than half that of adjacent conifers (Fechner and Barrows 1976). This might explain why wildfires spreading from high elevation conifer forests have been observed to die out in aspen. Healthy aspen stands are regarded by fire managers as relatively fireproof. It follows that maintenance and establishment of aspen are useful

---

[1] Department of Horticulture, Crop & Soil Sciences, New Mexico State University, Las Cruces, New Mexico. This paper published as New Mexico State Agricultural Experiment Station Scientific Paper No. 266. This research was funded by the Eisenhower Consortium and the Forest Service - USDA, grant number RM-81-160-GR. We are grateful to John B. McRae and Gregory Fancher for assistance.

fire management practices, particularly in mountain resort areas where ignition is likely and the potential for loss of resource value and life is great.

At present, land managers in the Southwest do not have an understanding of the steps necessary to grow aspen seedlings reliably and efficiently, or of those steps leading to fuelbreak establishment. Through a U.S. Forest Service-Eisenhower Consortium cooperative research project begun in 1981, we have developed or refined greenhouse, nursery, site preparation and weed control practices leading to aspen establishment. This paper reports the effects of aspen stock type on planting success.

METHODS

Two separate experiments were established, one in the Sacramento Mountains of south-central New Mexico in 1982, the other 1 year later in the Sangre de Cristo Mountains, 3 degrees latitude and 360 km to the north. The southern site (elevation 2,650 m) annually receives about 700 mm of precipitation, 550 mm of which are lost to evapotranspiration. The northern site (elevation 2,870 m) is more moist, averaging 900 mm precipitation and 400 mm evapotranspiration.

Aspen seeds were collected in early June 1981 from open-pollinated clones growing from 2500 to 2600 m elevation about 15 km northeast of Santa Fe, New Mexico. Seeds were processed by Harder's (1970) method and stored in the manner described by Benson and Harder (1972). Container seedlings were greenhouse grown for 17 weeks. Bareroot seedlings were produced in accordance with methods described by Benson and Einsphar (1962) and Benson and Dubey (1972). Stock types compared in the studies are described (Table 1).

Table 1. Description of four aspen stocktypes planted at two experimental sites.

Container Volume	Production Growing Density	Ht. (cm) $\bar{x}$ / range	Cal. (mm) $\bar{x}$ / range
164 cm^3	514/ m^2	34/ 28-40	4.0/ 2.2-4.4
350 cm^3	290/ m^2	45/ 35-60	4.1/ 2.2-4.5
870 cm^3	200/ m^2	55/ 45-65	4.8/ 3.6-5.2
Bareroot	130/ m^2	70/ 62-85	5.4/ 4.0-5.8

Sites were rototilled about 20 cm deep before planting with power augers. Seedlings were arranged in 3 m X 5 m, 40-tree rectangular plots in five replications at each site. Periodic hand weeding controlled weeds. Duplicate split-block experimental designs were used for both studies. At each site, main treatments tested May and July planting dates and subtreatments tested the stock types.

Survival and growth 1 year from planting were evaluated. At the southern site, only height growth was monitored, but in the north, stem caliper was also measured. Arcsine transformations were performed on the survival percentages before analysis (Little and Hills 1978). Treatment effects were compared with the protected LSD ($p \leq .05$). Relative growth rate (RGR) was calculated to determine changes in seedling height or caliper or both over time. A positive RGR reflects seedling growth relative to the seedling's size at planting. Negative RGRs indicate moderate to severe stem dieback with replacement by a resprout. RGR was calculated as follows:

$$RGR = \frac{m_2 - m_1}{m_1} \times \frac{1}{t_2 - t_1} \times 100$$

where

RGR = relative growth rate

m_1 = initial size at planting

m_2 = second measurement

t_1 = time at first measurement (in years)

t_2 = time at second measurement (in years)

RESULTS

Southern Site

Stock types were evaluated within each planting date (Fig. 1) because the planting time X stock type interaction was significant. One-year survival was higher for July-planted stock (54%) receiving late summer seasonal rains, than for May-planted stock (43%). Soil moisture to a depth of 30 cm was 30% at May planting but fell to 15% by mid June. Among May-planted trees, 73% of the bareroot seedlings were alive 1 year after planting, but survival among container stock types was uniformly poor, averaging 32%. For the July planting, survival was highest among 164-cm^3 and 350-cm^3 stock, and lowest among bareroot and 870-cm^3 container stock (Fig. 1). All stock types suffered stem dieback (Table 2) caused by drought. Because of dieback, trees averaged about 60% less height and caliper than when planted.

Fig. 1. One-year survival of four aspen stock types planted May and July, 1982 in the Sacramento Mountains, NM. Letters indicate significant differences within each planting date, as determined by the protected LSD ($p \leq .05$).

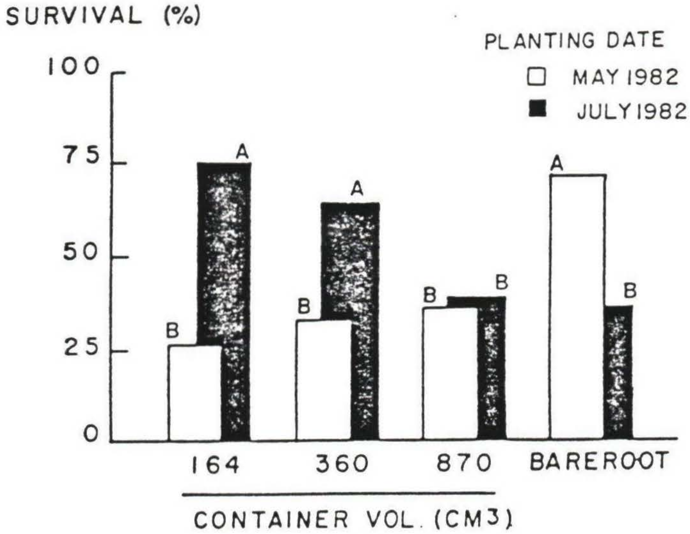


Table 2. Relative height growth (RHG) and relative caliper growth (RCG) of four aspen stocktypes 1 year from planting at two experimental sites.

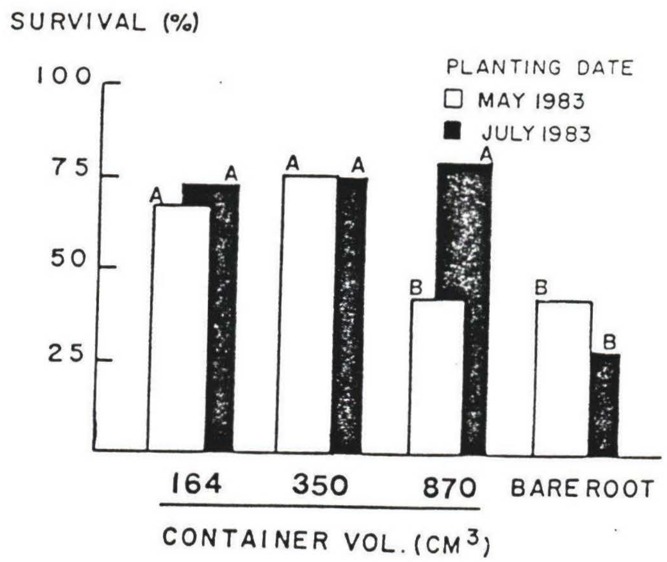
Stocktype	Southern Site		Northern Site			
	RHG (%/yr.)		RHG (%/yr.)		RCG (%/yr.)	
	May	July	May	July	May	July
Container						
164 cm^3	-58.2ab	-69.5c	12.6a	27.0a	37.2a	76.6c
350 cm^3	-72.1bc	-66.5bc	14.8a	23.6a	45.1a	105.5ab
870 cm^3	-76.6c	-48.2a	-22.9b	17.3a	17.2bc	81.0bc
Mean	-68.9	-61.3	1.44	22.6	33.1	87.7
Bareroot	-41.3a	-58.9abc	-39.9c	-65.8b	0.0c	-38.6d
Planting Date Mean	-62.0	-60.7	-8.88	0.48	24.8	56.0

Column values with the same letter are not significantly different ($p \leq .05$) as determined by protected LSD.

Northern Site

More moisture at this site yielded higher overall survival and growth than recorded for the southern site, although statistical comparisons between sites can not be made. Soil moisture to a depth of 30 cm did not fall below 24%. Overall survival was slightly higher for July-planted stock than May-planted stock (63% vs 57%). July-planted seedlings grew considerably better than May-planted stock (Table 2). Among May-planted stock, survival was highest for 164-cm^3 (68%) and 350-cm^3 stock (76%) and lowest for bareroot (42%) and 870-cm^3 container stock (42%) (Fig. 2). Among July-planted seedlings, survival across containers did not differ significantly (av. 74%) and was clearly superior to bareroot survival (27%). Container growth after July planting was also superior (Table 2). For the smaller container types, survival was similar for planting dates but more growth occurred after July planting.

Fig. 2. One-year survival of four aspen stock types planted May and July, 1983 in the Sangre de Cristo Mountains, NM. Letters indicate significant differences within each planting date, as determined by the protected LSD. ($p \leq .05$).



DISCUSSION AND CONCLUSIONS

Site and planting date influenced stock type performance. Survival across stock types planted at both sites indicates that the July planting date is superior to May. Specifically, survival and growth of container stock types were generally best when seedlings were planted in July rather than May. Among the 164-cm^3 and 350-cm^3 containers, July planting clearly improved survival at the southern site and growth at the northern site. May-planted bareroot stock performed satisfactorily at the southern site, apparently because transpirational surface area was minimal and growth was delayed during the May-June drought. However, in three of the four test plantings, survival among the smaller containers was considerably higher, averaging 70% or more. The 164-cm^3 container yielded greater than 75% survival on herbicide experimental sites found adjacent to stock type test areas (Fisher and Neumann, in press).

The overall more favorable first year response of the containerized stock corresponds with similar aspen field performance tests in Michigan (Okafo and Hanover 1978).

The stocktype recommended for establishing aspen in the southern rockies is the 164-cm^3 container. Survival of seedlings grown in 164-cm^3 containers was not exceeded by the larger containers produced at greater cost. The container seedlings used in this study bore fully expanded leaves. Possibly, container stock without leaves will have higher survival potential and this merits attention, particularly for planting droughty sites in early spring.

LITERATURE CITED

Benson, M.K. and D. Dubey. 1972. Aspen seedling production in a commercial nursery. Inst. Pap. Chem. Genet. and Physiology. Notes No. 12, 7 p.

Benson, M.K. and Einsphar. 1962. Improved method for nursery production of quaking aspen seedlings. Tree Planters' Notes No. 53:11-14.

Benson, M.K. and M.L. Harder. 1972. Storage of aspen seed. Inst. Pap. Chem. Genet. and Physiol. Notes No. 11, 4 p.

Fechner, G. H. and J. S. Barrows. 1976. Aspen stands as wildfire fuel breaks. Eisenhower Consortium Bull. 4, 26 p.

Fisher, J.T. and R.W. Neumann. Cultivation and herbicides promote aspen seedling establishment. In "Weed Control for Forest Productivity". Proc. of a meeting held Feb. 5-7, 1985, Spokane, Washington (in press).

Harder, M.L. 1970. Procedures for the collection and extraction of Populus seed. Instit. Pap. Chem. Genet. and Physiol. Notes. No. 9, 3 p.

Jones, J. R. and D. P. Trujillo. 1975. Development of some young aspen stands in Arizona. USDA For. Serv. Res. Pap. RM-151, 11 p.

Little, T. M. and F. J. Hills. 1978. Agricultural Experimentation Design and Analysis. John Wiley and Sons. New York. 350 p.

Okafo, O.A. and J.W. Hanover. 1978. Two year performance of barerooted and containerized trembling and bigtooth aspen seedlings. Tree Planter's Notes No. 29 (2):24-28.

Appendix Table 1. Comparison of production costs associated with each of the stock types tested in Exp. 6.

Stock type	container vol.	cost/ 100 surviving trees
Ray Leach tube	164 cm^3	11.50
CSU styroblock	350 cm^3	27.30
milk carton	870 cm^3	39.80
bare root	_	25.50

Costs include media, containers and energy. Bare root production costs are estimated from data on southern hardwood nursery production costs. Labor costs are not included.

Cost of surviving seedling calculated as follows:

Production Cost / average field survival

Average field survival includes May and July planting survival at northern and southern sites.

III. RESEARCH SUMMARY

The large-scale production of aspen planting stock from native seed is feasible, if nursery and greenhouse cultural procedures are rigorously followed. New Mexico aspen forests can be successfully regenerated from seedling stock. First year survival should exceed 75 %, if seedling stock and site management techniques steps are prescribed as suggested by the studies reported.

The availability of aspen seed in the southern Rockies was undocumented at the time these studies were begun. Over the course of the studies, several reliable seed sources were identified in northern New Mexico and southern Colorado. Seed production is variable from year to year, but appears adequate for sustaining the production of planting stock at the operational scale.

<u>Specific Findings</u>:

Results showed that the 68-in^3 (1115 cm^3) and 90-in.^{3} (1475 cm^3) containers produced seedlings with greater shoot and root dry weights, root collar caliper, and height than did 11-in^3 (180 cm^3) or 30-in.^{3} (492 cm^3) containers. Because the 68-in^3 container uses 24% less media than the 90 -in^3, the 68-in^3 unit is considered the best for production of large container stock. However, in subsequent field planting tests the 10-in^3 (164 cm^3) container proved most satisfactory. The growth rates derived for the four container sizes can be used to schedule production.

Plantable aspen nursery seedlings can be successfully grown on moderately alkaline, southwestern mountain valley sites if the nursery bed is amended with peat moss and sulphur. Peat moss markedly improved soil medium physical and chemical properties responsible for improving seedling growth. Sulphur alone did not produce satisfactory growth. The necessity of sulphur in the presence of peat was not tested.

Cupric carbonate treatments did not improve container seedling transplant quality. That is, $CuCO_3$ did not positively alter root fibrosity. Also, in contrast to results reported for conifers, container wall chemical treatments did not improve aspen seedling growth above controls and often adversely affected growth.

Forest soil provides an effective means for assuring the production of mycorrhizal aspen seedlings in containers. Inoculation at the 8 and 16 % (v/v) levels significantly increased intensity of mycorrhizal infection and seedling biomass over controls (not inoculated).

Ectomycorrhizae formed under the inoculation treatments were morphologically similar to types previously reported for aspen. No endo types were detected, even when a proven source of VAM inoculum was applied in the absence of fertilizer phosphorus. This suggests that aspen may depend predominantly on ecto types. Because no endo symbionts were detected, results offer few insights into the potential role of endo symbionts in improving seedling quality. Because greenhouse seedlings were grown at fertilization levels considered below optimum for rapid growth, the ideal balance between mycorrhizal infection and biomass accretion remains a problem to be researched.

First year survival exceeded 75% for the best site preparation treatment at each site. Cultivation, in general, effectively reduced weed cover and improved seedling success. Fall cultivation, in particular, improved seedling survival and growth only at the relatively dry southern site. Except for spring-cultivated plots in the south, some herbicide applications improved weed control and seedling performance over cultivation alone. The combination of fall cultivation plus trifluralin is considered the best site preparatory treatment tested.

Site and planting date influenced stocktype performance. Survival across stock types planted at both sites indicates that July planting date is superior to May. Specifically, survival and growth of container stock types were generally best when seedlings were planted in July rather than May. Among the 164-cm^3 (10-$in.^3$) and 350-cm^3 (21.4-$in.^3$) containers, July planting clearly improved survival at the southern site and growth at the northern site. May-planted bareroot stock performed satisfactorily at the southern site, apparently because transpirational surface area was minimal and growth was delayed during the May-June drought. However, in three of the four test plantings survival among the smaller containers was considerably higher, averaging 70 % or more. The 164-cm^3 container yielded greater than 75 % survival on herbicide experimental sites found adjacent to stock type test areas. The overall more favorable first year response of the containerized stock corresponds with similar aspen field performance tests in Michigan (Okafo and Hanover 1978).

The stock type recommended for establishing aspen in the southern rockies is the 164-cm^3 container. Survival of seedlings grown in 164-cm^3 containers was not exceeded by the larger container produced at greater cost. (Cost of surviving seedling is presented in Appendix Table 1.) The container seedlings used in this study bore fully expanded leaves. Possibly, container stock without leaves will have higher survival

potential and this merits attention, particularly for planting droughty sites in early spring.